Alternative Routes of Polysaccharide Acylation:
Synthesis, Structural Analysis, Properties

Dissertation

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Abbreviations

AGU	Anhydroglucose unit
AFM	Atomic-forced microscopy
CA	Cellulose acetate
CAP	Cellulose acetate propionate
CDI	*N*,*N*`-Carbonyldiimidazole
CE	Cellulose esters
CTA	Cellulose triacetate
CMC	Critical micelle concentration
DCC	*N*,*N*-Dicyclohexylcarbodiimide
DMAc	*N*,*N*-Dimethylacetamide
DMAP	4-Dimethylamino pyridine
DMF	*N*,*N*-Dimethylformamide
DP	Degree of polymerisation
DS	Degree of substitution
DS_{EA}	Degree of substitution calculated by EA
DS_{Tit}	Degree of substitution calculated by titration method
EA	Elemental analysis
Fig.	Figure
FTIR	Fourier-transform infrared spectroscopy
GPC	Gel permeation chromatography
HPC	Hydroxypopyl cellulose
HPLC	High performance liquid chromatography
LB	Langmuir-blodgett
NMR	Nuclear magnetic resonance
NS	Number of scans
PP	4-Pyrollidinopyridine
SAMs	Self-assembled monolayers
SPR	Surface plasmon resonance
Tab.	Table
TBAF	Tetrabutylammonium fluoride trihydrate
*T*d	Thermal decomposition temperatures
TFA	Trifluoroacetic acid

TGA	Thermogravimetric analysis
THF	Tetrahydrofuran
TMSC	Trimethylsilylated cellulose
Tos-Cl	*p*-Toluenesulfonyl chloride
TosOH	*p*-Toluenesulphonic acid
Tosyl	*p*-Toluenesulfonyl
ν	Wave length measured in cm^{-1}
δ	Chemical shift in ppm

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1. Introduction

1.1. General aspects and structural features of cellulose

Cellulose constitutes the most abundant renewable polymer resource available today worldwide. Payen recognized cellulose as a definitive substance and coined the name "cellulose" (Payen, 1838). Cellulose as a precursor for chemical modification has been used even before its polymeric nature was recognized and well understood. Milestones on this pathway were the discovery of cellulose nitrate (Schönbein, 1846), the preparation of Schweizer's reagent, i.e cuprammonium hydroxide solution representing the first cellulose solvent (Schweizer, 1857) and synthesis of an organo-soluble cellulose acetate by Schützenberger in 1865 (Schützenberger, 1865, 1865a). The origin of cellulose chemistry as a branch of polymer research can be traced back to the fundamental experiments of H. Staudinger in the 1920's and 1930's on the acetylation and deacetylation of cellulose; these experiments resulted in the concept of polymer-analagous reactions (Staudinger and Daumiller, 1937). Regarding source of cellulose, plant/wood is the major source while cotton is the best source of highly pure cellulose. The chemosynthesis of functionalized cellulose has been experimentally realized (Nakatsubo et al., 1996; Nishimura et al., 1993). The non-biosynthesis with controlled molecular weight preparation of cellulose was described involving an enzymic polymerization (Kobayashi et al., 1991).

Further cellulose products like methyl-, ethyl-, or hydroxyalkyl ethers or cellulose acetate, and, in addition, products with combinations of various functional groups, e.g. ethylhydroxyethyl cellulose, hydroxypropylmethyl cellulose, cellulose acetopropionates and cellulose acetobutyrates are still important, many decades after their discovery. Also ionic cellulose derivatives are known for a long time. Carboxymethyl cellulose, up to know the most important ionic cellulose ether, was first prepared in 1918 and was produced commercially in the early 1920's in Germany (Balser et al., 1986). These are produced in large quantities for diversified applications.

Cellulose is a polydisperse linear homopolymer consisting of regio- and enantioselective β-1, 4-glycosidic linked D-glucose units (AGU, Fig.1.1). The polymer contains three hydroxyl groups at the C-2, C-3 and C-6 atoms, which are, in general, accessible to the typical conversions of primary and secondary alcoholic OH groups. In this context, it is worth mentioning that the vicinal secondary hydroxyl groups may undergo typical glycol reactions. Based on this molecular structure, i.e. the "tacticity" and the uniform distribution of the hydroxyl groups, ordered hydrogen bond systems from various types of supramolecular semicrystalline structures (Krassig, 1993; Salmon et al., 1997). The significance of the accessibility factor in affecting the cellulose reactivity in generally accepted today. Not only the crystallinity but also hydrogen bonding patterns has a strong influence on the whole chemical behavior of cellulose (Lai, 1996). A further consequence of the supramolecular structure as the insolubility of the macromolecule in water as well as in common organic liquids which stimulated and still stimulates the search for solvents appropriate for homogeneous phase reactions which are still unconventional synthesis paths.

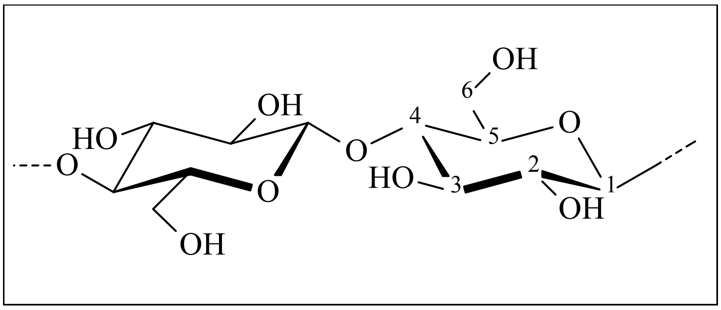


Fig. 1.1: Structure of cellulose

1.2. Aims and objectives

The aim of the research was to study novel synthetic methods of cellulose esterification, characterization of structures and properties of the esters synthesized.

The interest was to prepare tailored cellulose esters via *in situ* activation of the carboxylic acids with Tos-Cl. The effects of base on DS and DP were studied. Effects of change in molar ratio and reaction times on the DS of the cellulose esters of long chain aliphatic carboxylic acids were examined. Some properties including thermal stability and solubility of the esters were examined as well. Interest was to synthesize homogeneously organo-soluble cellulose lipoates for film formation over gold.

Novel and very efficient reagent iminium chloride was explored for the *in situ* activation of carboxylic acids to functionalize cellulose. Further, this new method is utilized for the synthesis of aliphatic, aromatic and bulky carboxylic acid esters of cellulose. Reaction mechanism of the iminium chloride formation of the carboxylic acid was studied.

Interest was developed to use novel soft and efficient acylating agent CDI in the novel solvent system DMSO/TBAF. This new method yielded products with high purity and less degradation of the cellulose backbone. Different carboxylic acids were possible to activate with CDI and to prepare esters in DMSO/TBAF solvent system. Reaction mechanism of the imidazolide formation of the carboxylic acid was studied.

Cellulose present in wood is closely associated with the hemicelluloses (pullulan) and lignins. In one project, aim was to determine the adsorption properties of pullulan onto regenerated cellulose surface. As stated above, pullulan has the ability to self-assemble in solution and therefore it can aid in the study of hemicellulose/cellulose interactions. Pullulan derivatives that contain abietic acid will also be studied to determine their effect on adsorption behavior.

Interest was also focused to use above synthetic methods of cellulose esterification for the synthesis of abietates of structurally related polysaccharides like hydroxypropyl cellulose and dextran.

Interest was focused to study different synthesis paths and analytical methods of cellulose esterification regarding structure elucidation of cellulose esters, especially cellulose acetates. The cellulose acetates were analyzed using ^{1}H NMR spectroscopy.

1.3. Literature

1.3.1. New analytical tools for structural elucidation of cellulose esters

Cellulose acetate (CA) is commercially produced since decades and has gained special technical importance due to its wide spectrum of properties as bio-based material. CA is serving humanity from a century since it was synthesized first. Chemical structure of CA is established well and several new synthesis paths appeared for its synthesis in lab or at industrial scale.

To establish structure-property relationships of CA and to evaluate synthesis paths and products, a detailed structure analysis is an unambiguous prerequisite. A broad variety of spectroscopic and chromatographic methods were investigated towards their use as analytical tool for the structure elucidation of CA. The most convenient method for the elucidation of structural features of CA is IR spectroscopy. In recent years attempts were made to use this method for a quantitative evaluation of the amount of bound acetic acid and the distribution of the primary and secondary hydroxyl groups in highly substituted CA samples.

The application of NMR spectroscopy was among the first attempts for the structure elucidation of CA. The pioneering work of both Goodlett (Goodlett et al., 1971) using ^{1}H NMR spectroscopy and Kamide (Kamide, 1981) applying ^{13}C NMR spectroscopic measurements opened major routes for further studies in this field including complete signal assignment, the determination of the functionalization pattern of CA dependent on reaction conditions and the establishment of structure-property relationships. However, the exact distribution of substituent in CA over a wide range of degree of substitution (DS) is not readily estimated by simple comparison of the relevant peak intensities. A major problem is the overlapping of signals around 70-85 ppm resulting of the unmodified C-2, 3, 5. In addition line broadening of the signals due to the ring carbons is frequently observed in quantitative mode of ^{13}C NMR measurements.

Attempts were made to exploit ^{1}H NMR spectroscopy for the structure determination. It was possible to calculate the partial DS at the free reactive sites from ^{1}H NMR spectra after peracetylation of the CA derivatives with acetyl-d_3-chrolide or acetic anhydride-d_6. The DS can be readily calculated from the ratio of the spectral integrals of protons of repeating unit and the methyl protons. The error of calculations increases due to the degrees of absolute signal intensity in case of samples of rather low DS.

One important alternative to NMR spectroscopy is the determination of the inverse substitution pattern of the hydrolytically unstable cellulose esters by means of chromatographic techniques after subsequent functionalization and depolymerisation. Among the first attempts in this regard was a method developed by Björndal (Björndal et al., 1971).

^{1}H NMR spectroscopy is being exploited to determine structure analysis of CA 2.5 after completele functionalization of free OH groups of CA 2.5 by using different synthetic methods, i.e. acetylation, propionylation, trifluoroacetylation, nitrobenzoylation, phenylcarbanilation and conversion with aceticacidethylester isocyanate. Especially the later one is a very efficient tool, which can’t only be applied for structure elucidation by means of NMR spectroscopy but also for HPLC studies after polymer degradation.

1.3.2. Unconventional cellulose esterification with carboxylic acids

Fast growing interests to prepare tailored derivatives of cellulose e.g. to prepare membranes for proteins filtration, thin films over gold and use in sensors stimulate the search for unconventional synthetic pathways for cellulose modification. Esterification of cellulose under homogeneous reaction conditions provides access to a variety of bio-based materials with valuable properties (Sealey et al., 1996; Heinze and Liebert, 2001; Heinze et al., 2003).

Reaction rate and final DS in heterogeneous reactions are hindered by low accessibility of solid cellulose to the esterification reagents (Sealey et al., 1996). Only a limited number of cellulose esters and ethers, mainly prepared under heterogeneous reaction conditions, found commercial interest although cellulose derivatives are known for over one century now. A major stimulation for the synthesis of polysaccharide esters was the development of new solvents and the investigation of *in situ* activation methods for carboxylic acids (Heinze and Liebert, 2001; Heinze et al., 2003). These tools can overcome the major disadvantages of the heterogeneous esterification with acid chlorides and anhydrides e.g. uncontrolled functionalization and side reactions, limited commercial availability or high costs, and time consuming purification of the products.

Cellulose esters of C2-C4 carboxylic acids including mixed products represent a class of commercially important polymers with excellent fibre and film forming characteristics. They have gained technical importance because of a wide spectrum of properties (Müller and Leuschke, 1996). The commercial production of the cellulose ester is exclusively carried out by the conversion of the polymer with acid anhydrides in the presence of mineral acid like H_2SO_4 as a catalyst. It is difficult to prepare esters of higher carboxylic acids (>C4) by this method due to the slow reaction rate and competitive cellulose chain cleavage. In case of carboxylic acid anhydrides, only half of the reagent incorporates in the product whereas the other part is converted into the corresponding carboxylic acid. Malm described a useful approach to the preparation of long chain cellulose esters (Malm et al., 1951). The reaction of cellulose with acid chloride in 1,4-dioxane and pyridine as an acid acceptor and catalyst afforded cellulose triesters ranging from acetate to palmitate. This method works quite well for the synthesis of products with a high DS. It requires amorphous, reactive regenerated cellulose as starting material. Kwatra described a novel synthesis method for cellulose esters with long chain palmitic acid (Kwatra et al., 1992). It involves the reaction of mercerised cellulose with the acid chloride at elevated temperature under vacuum to facilitate the removal of the by-product HCl.

Recently, alternative paths of acylation of cellulose starting from the dissolved polymer and/or with the application of special reagents are important research interests in academics (Heinze and Liebert, 2001). The most versatile and interesting types of cellulose solvents for the modification are binary mixtures of polar organic liquids and inorganic salts (Morooka et al., 1984; Philipp et al., 1986; Klemm et al., 1998). Typical examples are DMAc or its cyclic analogue N-methyl pyrrolidone in combination with LiCl (Samaranayake and Glasser, 1993). The most important solvent of cellulose with regard to homogeneous esterification is DMAc in combination with LiCl. Accessibility of cellulose to reactant increases in solution form and homogeneous reaction needs milder conditions than analogous heterogeneous reaction (Sealey et al., 1996). The esterification of cellulose in DMAc/LiCl was extensively studied during the last decade (Dawsey 1994; El Seoud et al. 2000). It was shown that cellulose can be reacted with carboxylic acid anhydrides, acid chlorides and other electrophilic acyl derivatives using mineral acid or alkaline catalyst (amines) to afford partially substituted cellulose esters with a uniform distribution of the functional groups (Diamantoglou and Kuhne, 1988; Samaranayake and Glasser, 1993 and 1993a; Glasser et al., 1995, Sealey et al., 1996). The synthesis of highly reactive acid chlorides is difficult especially if the carboxylic acids contain sensitive moieties and, moreover, acid chlorides are mostly insoluble in DMAc/LiCl in the presence of triethylamine as base.

Recently, the mixture DMSO/TBAF was found to be a very efficient solvent for cellulose, which is increasingly studied as reaction medium. DMSO/TBAF dissolves cellulose without any pre-treatment within 15 minutes. It has been exploited for acylation reactions using acid anhydrides and vinyl esters (Heinze et al., 2000; Ciacco et al., 2003; Heinze and Liebert, 2001).

An interesting new path is the *in situ* formation of reactive carboxylic acid derivatives. Tos-Cl (Shimuzu and Hayashi, 1988; Gräbner et al., 2002; Sealey et al., 1996; Glasser et al., 2000; Heinze and Schaller, 2000) and *N,N*-dicyclohexylcarbodiimide (DCC) in combination with 4-pyrollidinopyridine (PP) (Samaranayake and Glasser, 1993; McCormick and Dawsey, 1990) were investigated extensively for the *in situ* activation of carboxylic acids and the conversion of cellulose dissolved in DMAc/LiCl. Tosyl chloride is a very efficient reagent. In contrast to older references assuming the mixed anhydrides as intermediate it was found recently that during the reaction the acid chloride and the symmetric anhydride are formed resulting in the high reactivity but remarkable side reactions e.g. degradation of the polymer (Heinze et al., 2003). It was shown that cellulose esters having alkyl substituents in the range form C12 to C20, can be obtained with almost complete functionalization of the accessible OH groups (Sealey et al., 1996). A variety of different cellulose esters was successfully synthesised via this path (Koschella et al., 1997; Heinze, 1998; Heinze et al., 2000; Heinze and Schaller, 2000). DCC/PP was successfully applied for preparation of long chain fatty acid esters (up to C-20) with complete functionalization of all the hydroxyl groups. Disadvantages of this method arise from the high toxicity of the reagent and the necessary work up (Samaranayake and Glasser, 1993).

An equally mild and efficient method is the *in situ* activation of carboxylic acids via iminium chlorides. They are simply formed by conversion of DMF with a variety of chlorinating agents including phosphoryl chloride, phosphorus trichloride or oxalyl chloride (Stadler, 1978). It was possible to isolate these hydrolytically instable intermediates (Feher and Stadler, 1975). Esterification of cellulose via the iminium chlorides of carboxylic acids of different substructures, i.e. acetic acid, the long chain aliphatic acids stearic acid and palmitic acid, the aromatic acid 4-nitrobenzoic acid and adamantane 1-carboxylic acid as bulky alicyclic acid, is the topic of interest and being reported for the first time.

CDI as activating agent for carboxylic acids has been used for the first time for the homogeneous cellulose modification in DMAc/LiCl (Gräbner et al., 2002). The advantages are milder reaction conditions, limited amounts of by-products, i.e. CO_2, which are non-toxic and reusable (Staab, 1962) and the commercial availability of the reagents. There is no reference found for the synthesis of cellulose lipoate even using CDI as activating agent and DMAc/LiCl as solvent system. Nothing is found in literature for the thin films formation of cellulose lipoate. We are reporting for the first time synthesis of cellulose lipoates and its self-assembly behavior onto gold surface.

Self-assembled thin films have become a well-established field. Thin films are mechanically and solvolytically stable. They can serve as model systems to study fundamental interfacial properties (Charych et al., 1992), such as wetting (Abbott et al., 1995; Nuzzo et al., 1990), friction (Depalma et al., 1989), adhesion (Ferguson et al., 1991), pattern definition (Kumar et al., 1994) and biomineralization (Küther et al., 1998). Systematic alteration of monolayer, in terms of chain length or functionality of the terminal groups, can also be carried out to study the influence on crystal growth. Rodziguez has studied the growth of hydroxyapatite crystals on cellulose matrix using titanium alkoxide as a coupling agent (Gonzalez et al., 2003). Cellulose derivatives can also be employed to study the enzyme immobilization on surfaces (Rebelo et al., 1997). Tanaka has demonstrated that thin films (5-10 nm) of regenerated cellulose could serve as ideal inter layers to deposit model and native cell membranes (Rehfeldt and Tanaka, 2003).

An efficient and mild homogeneous synthesis of pure aliphatic, alicyclic, aromatic, and bulky carboxylic acid esters of cellulose using CDI as activating agent in DMSO/TBAF solvent system is being studied. Besides synthesis, investigation for reaction mechanism of carboxylic acid imidazolide formation with the help of NMR spectroscopy is also topic of work.

1.3.3. Synthesis and self-assembly behavior of amphiphilic pullulan abietates

Pullulan, first described in 1959, (Wallenfells et al., 1961) is a water soluble extracellular polysaccharide produced by strains of *Aureobasidium pullulans* (Youssef et al., 1999; Lazaridou et al., 2002) consisting of a linear and flexible chain of D-glucopyranosyl units that alternate regularly between one α-(1,6) and two α-(1,4) linkages (Muroga et al., 1987). Owing to its oxygen impermeability, non-toxic and non-irritating properties, it is used for producing films, binders, adhesives, thickners, viscosity improvers and coating agents. Thus, pullulan has a number of potential uses in the pharmaceutical and food industries and in other fields of biotechnology. By introducing functional groups into the pullulan macromolecule, it is possible to improve its performance and extend the fields of possible applications.

Chemical modification of pullulan may be performed, as with cellulose, by esterification or etherification of hydroxyl groups in a maltotriosyl unit. As the maltotriosyl unit contains 9 hydroxyl groups in a geometrically unique environment, the structural diversity of pullulan derivatives surpasses that in cellulose or other polysaccharides having a single glycosyl repeating unit (Tezuka et al., 1998).

A number of publications and, especially, patents discuss a variety of pullulan derivatives and their potential applications: chlorinated (Mayer, 1990), chloroalkylated (Mocanu et al., 1992, 1999), sulphinylethylated (Imai et al., 1991) etherified (Fujita et al., 1978; Nishijima et al., 1979), cyanoethylated (Onda et al., 1981; Murase et al., 1983), carboxylated (Tsuji et al., 1976), permethylated (Keilich et al., 1971), cationized (Onishi, 1985), sulphated (Carpov et al., 1985), acetylated (Hijiya et al., 1974, 1974a, 1975), esterified (Hijiya et al., 1974a). Solvents used for esterification reactions were DMF, DMAc, *N*-methyl pyrrolidone. Tezuka recently performed synthesis of pullulan nonaacetate using acetic anhydride in pyridine and DMAP (Tezuka, 1998).

In literature no reference was found to synthesize abietic acid esters of pullulan. Synthesis of abietic acid esters of pullulan in DMAc solvent using differently activated abietic acid, i.e. *in situ* activation with Tos-Cl, CDI, and oxalyl chloride is being studied for the first time. Abietic acid is a hydrophobic molecule extracted from tree resin (Hillis et al., 1962).

Wood is one of nature's most fascinating materials yet to be mimicked synthetically. Through the study of self-assembly behavior of pullulan onto a model cellulose surface give further insight into the interactions between a hemicellulose and cellulose. Pullulan is hydrophilic, but if pendant side chains containing hydrophobic groups are attached, amphiphilic character is established (Uraki et al., 1997). Akiyoshi has also studied the self-assembly behavior of hydrophobized polysaccharides in water (Akihiro et al., 1978; Akiyoshi et al., 1993). Akiyoshi found that cholesterol substituted pullulan derivatives were capable of forming hydrogel nano-particles by their self-assembly in water.

2. Results and Discussions

2.1. Chemical characterization of cellulose esters

To establish structure-property relationships of cellulose esters (CE) and to evaluate synthesis paths and products, a detailed structure analysis is an unambiguous prerequisite. The application of NMR spectroscopy was among the first attempts for the structure elucidation of CA (Goodlett et al., 1971). Attempts were made to exploit ^{1}H NMR spectroscopy for the structure determination. Different alternatives for a subsequent derivation of CE for analytical purposes were developed, which is essential to calculate degree of substitution (DS) of CE using ^{1}H NMR spectroscopy. In this work mainly ^{1}H NMR spectroscopy was explored to determine detailed structure of CE exemplified for cellulose acetate (CA).

2.1.1. Propionylation of CA 2.5

In order to get well-resolved ^{1}H NMR spectrum, CA 2.5 was propionylated using propionic anhydride in pyridine as solvent. Complete functionalization of CA was possible. Experiment was carried out in duplicate and ^{1}H NMR spectra of CAP (sample **CA-1**) synthesized were also recorded twice. ^{1}H NMR spectrum of CAP is shown in Fig. 2.1.

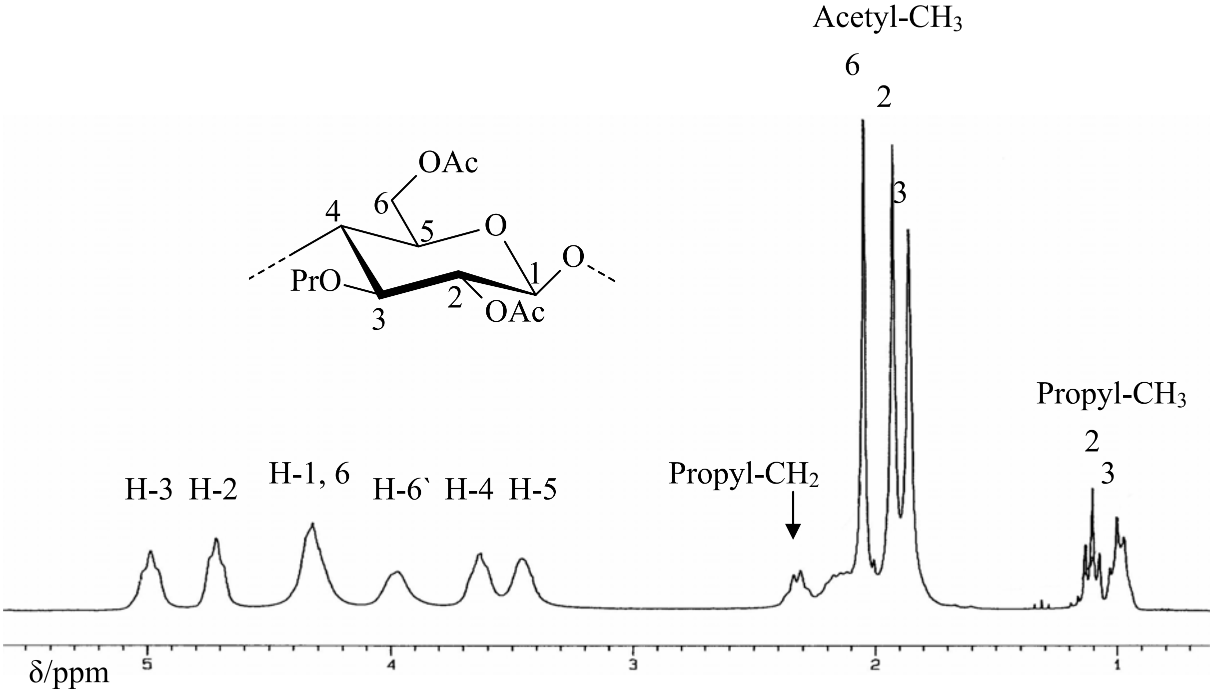


Fig. 2.1: ^{1}H NMR (CDCl$_3$, NS 32) spectrum of cellulose acetate propionate **CA-1**

DS_{Acetyl} 2.32, 2.32,2.35,2.35 and 2.37, 2.37,2.38,2.38 were found ($S^2 = 1.32 \times 10^{-4}$) for acetyl moities. DS_{Acetyl} calculated by the ratio of spectral integrals of acetyl moiety and repeating unit. It was thought that trans-esterification occurred when perpropionylation of CA is carried out using propionic anhydride. For this purpose CA 2.5 was peracetylated completely in homogeneous reaction medium using acetic anhydride and pyridine yielded product **CA-2**. A typical ^1H NMR spectrum of CTA (**CA-2**) is shown in Fig. 2.2. Spectrum indicated complete conversion of the CA 2.5 to CTA (DS 2.96). Three signals of methyl protons appeared which are assigned to the acetyl moieties at position 2, 3 and 6. Total DS of acetyl moieties DS = 2.96, which was calculated from the ratio of the spectral integrals of protons of repeating unit and the methyl protons. CTA was allowed to react with propionic anhydride at 60°C and 120°C and ^1H NMR spectra were recorded. DS_{Acetyl} was calculated is 2.96 in both cases, which is equal to the DS of CTA we used for this reaction. Moreover no signal of propyl moiety appeared in ^1H NMR spectra, which exclude the doubt of trans-esterification.

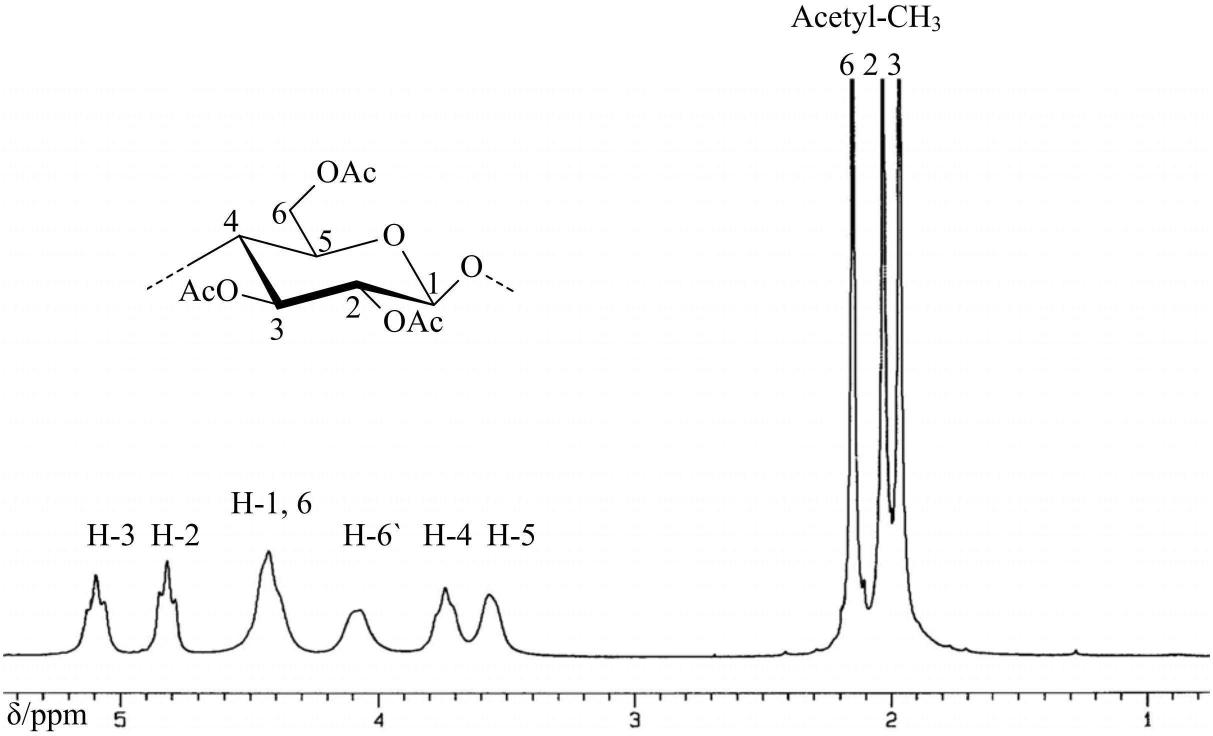


Fig. 2.2: ^1H NMR ($CDCl_3$, NS 16) spectrum of cellulose triacetate **CA-2**

2.1.2. Trifluoroacetylation of CA 2.5

Trifluoroacetylation of CA 2.5 was carried out using CDI as *in situ* activating agent. CDI reacts with TFA in DMSO to yield imidazolide of TFA, which reacts with the free hydroxyl functions of CA. By this method, it was not possible to completely substitute the free OH groups of CA by using 1:3:3 mole ratios of CA 2.5/CDI/TFA at 80°C. However, acetone soluble product was obtained, which yielded ^{1}H NMR spectrum better resolved then CA 2.5 showing introduction of TFA moiety (sample **CA-3**, see Fig. 2.3). ^{1}H NMR spectrum (acetone-d_6) showing three signals of methyl protons, which are assigned to the acetyl moieties at position 6, 2 and 3. Less resolved AGU appeared in the range of 3.5-5.1 ppm.

The reason of incomplete functionalization of CA 2.5 using TFA/CDI might be the instability of TFA in water/moisture in the system. Another explanation might be possible that the imidazolide of strong carboxylic acid (TFA) may leads to less reactive imidazolide. Similar results for acetylation of Avicel cellulose with acetic acid and CDI were observed in other experiments by which we could not get completely functionalised and organo-soluble CTA.

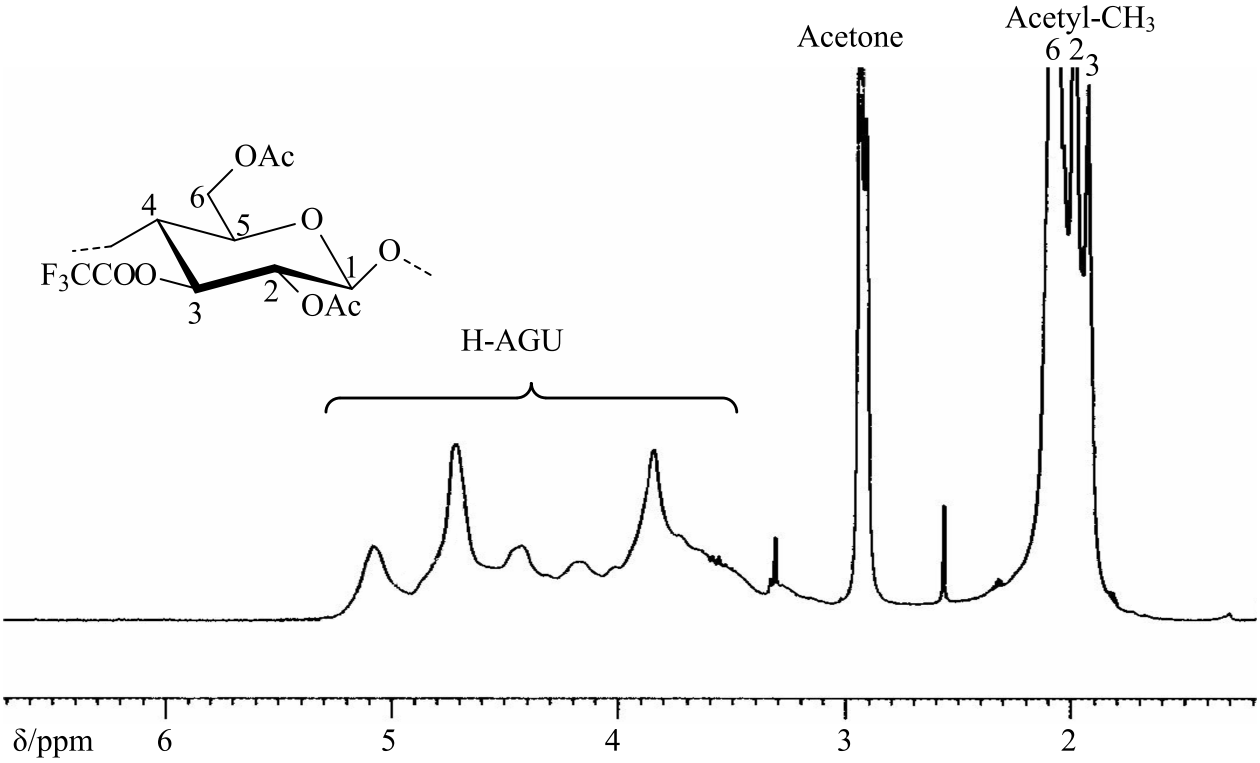


Fig. 2.3: ^{1}H NMR (acetone-d_6, NS 16) spectrum of cellulose acetate trifluoroacetate **CA-3**

2.1.3. Nitrobenzoylation of CA 2.5

A valuable tool for the analysis of CA is nitrobenzoylation of remaining OH groups of CA 2.5. Nitrobenzoylation of CA was carried out homogeneously using imidazolide of 4-nitrobenzoic acid, which can be prepared easily by reacting 4-nitrobenzoic acid with CDI in DMSO at room temperature. Re-precipitated CA 2.5 from THF into ethanol was used in order to avoid acetic acid imidazolide formation during the reaction because acetic acid is present as impurity in the CA 2.5. Complete substitution of the remaining OH groups of CA was possible by using 6 times of reagents (4-nitrobenzoic acid/CDI) to free hydroxyl of CA 2.5, i.e. 1:3:3 molar ratios (unsubstituted AGU/CDI/4-nitrobenzoic acid) at 80°C. DS_{Acetyl} 2.60 was calculated from the ratio of spectral integrals of repeating unit and aromatic protons of 4-nitrobenzoate moieties. ^1H NMR spectrum of cellulose acetate 4-nitrobenzoate **CA-4** was recorded in $CDCl_3$ is shown in Fig. 2.4.

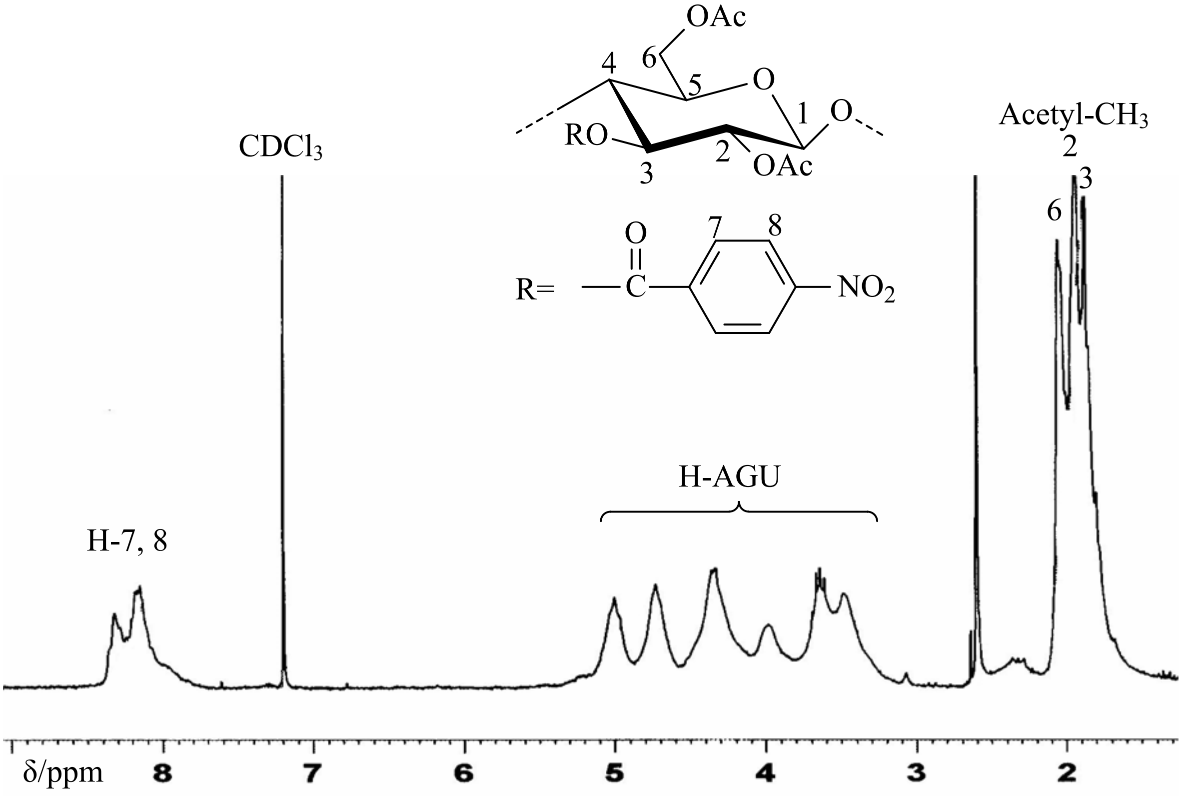


Fig. 2.4: ^1H NMR ($CDCl_3$, NS 16) spectrum of cellulose acetate 4-nitrobenzoate **CA-4** synthesised via imidazolide formation

Another homogeneous synthetic path for nitrobenzoylation was studied using 4-nitrobenzyl chloride in pyridine. Complete substitution of the remaining OH groups of CA was possible by using 1:3:3 molar ratios (unsubstituted AGU/CDI/4-nitrobenzoic acid) of reactants, which yielded product **CA-5**. ^{1}H NMR spectrum of cellulose acetate 4-nitrobenzoate **CA-5** was recorded in $CDCl_3$ is shown in Fig. 2.5. DS_{Acetyl} 2.66 was calculated from the ratio of spectral integrals of repeating unit and aromatic protons of 4-nitrobenzoate moieties, which was comparable to the DS_{Acetyl} 2.60 calculated after nitrobenzoylation via above described method using 4-nitrobenzoic acid/CDI. Both the methods yielded similar ^{1}H NMR spectra showing well-resolved AGU at 3.5-5.1 ppm. Spectrum showed three signals of methyl protons assigned to the acetyl moieties at position 2, 3 and 6. Aromatic protons of nitrobenzoate moieties appeared at 7.92-8.45 ppm.

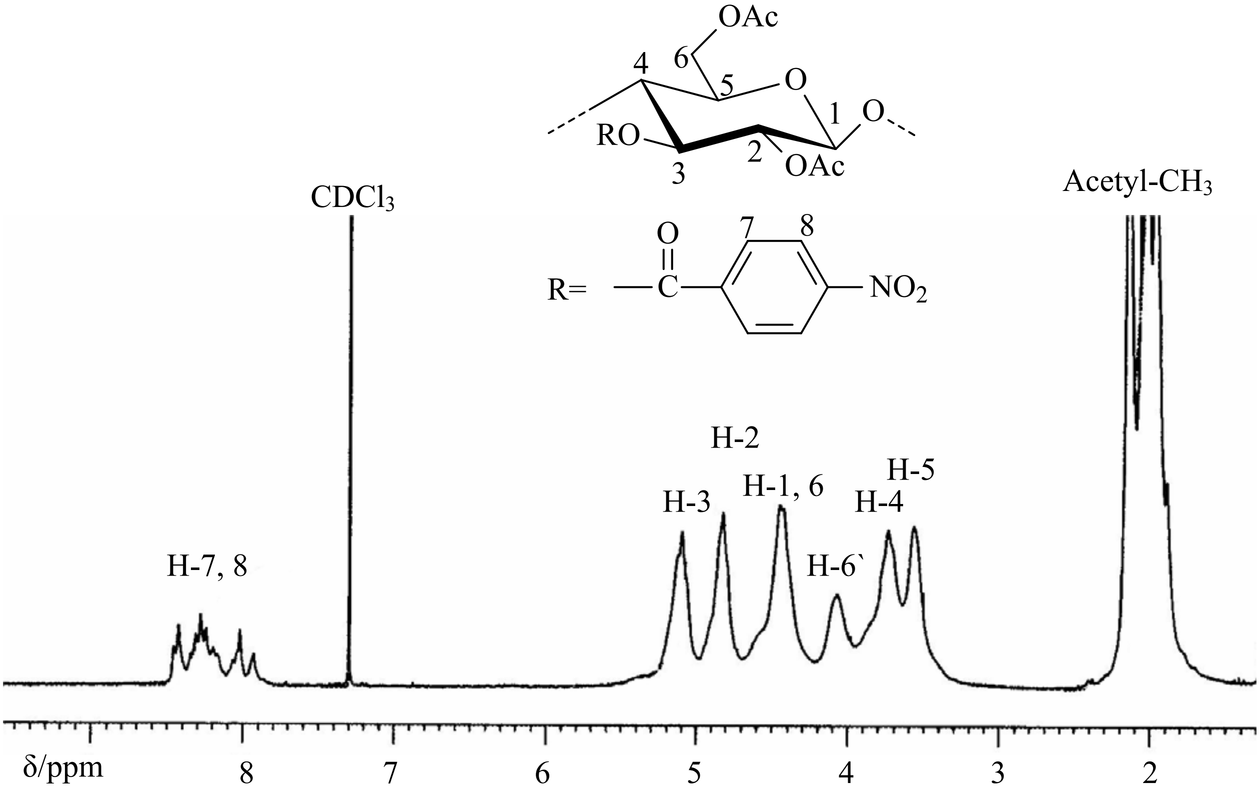


Fig. 2.5: ^{1}H NMR ($CDCl_3$, NS 16) spectrum of cellulose acetate 4-nitrobenzoate **CA-5** synthesised with 4-nitrobenzyl chloride

2.1.4. Conversion of CA 2.5 with ethylisocyanato acetate

The functionalization of CA 2.5 was carried out by reacting CA 2.5 with ethylisocyanato acetate. In this reaction path, CA 2.5 dissolved in pyridine was allowed to react with ethylisocyanato acetate at 100°C. The completely substituted CA-ethylcarbamate **CA-6** was formed, which is chloroform soluble product. ^{1}H NMR spectrum of **CA-6** was recorded in chloroform showing well-resolved AGU (Fig. 2.6). Spectrum showed three signals of methyl protons assigned to the acetyl moieties at position 6, 2 and 3 respectively at 2.18, 2.02 and 1.91 ppm. Carbamate-CH_3 protons appeared at 1.29 ppm. DS_{Acetyl} 2.43 was calculated from the ratio of spectral integrals of repeating unit and methyl carbamate protons.

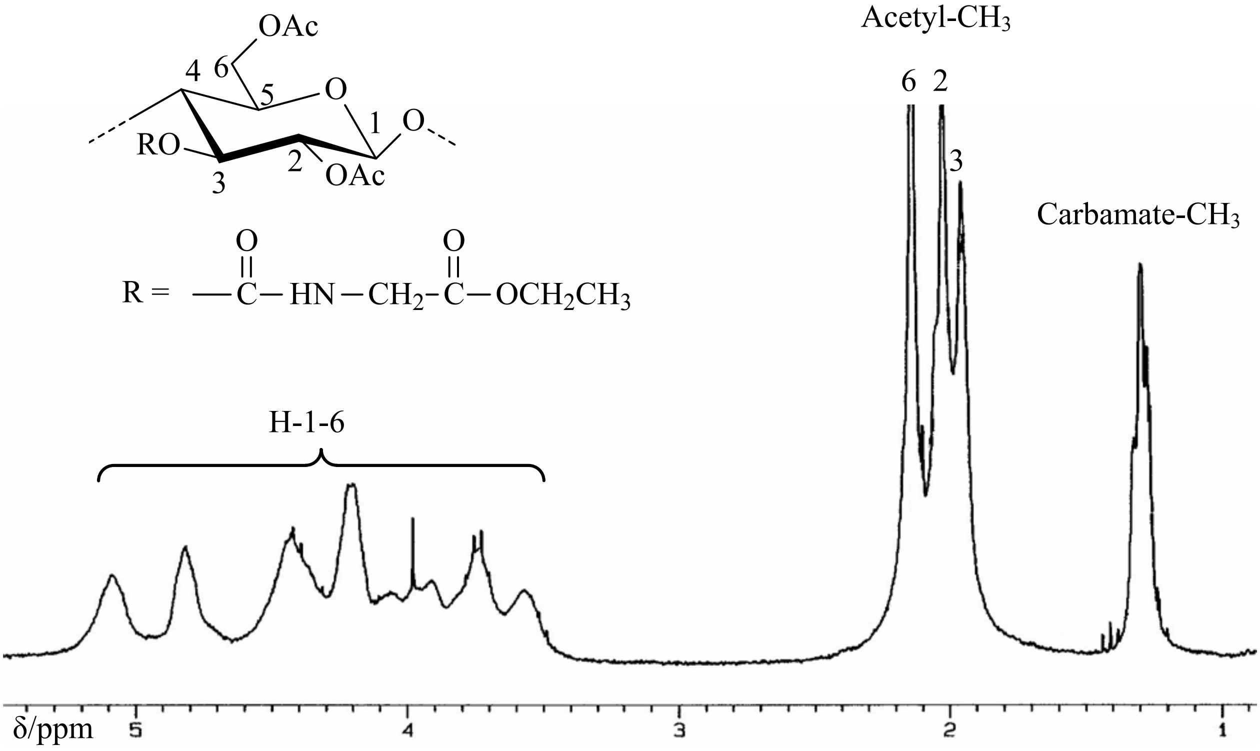


Fig. 2.6: ^{1}H NMR ($CDCl_3$, NS 16) spectrum of cellulose acetate ethylcarbamate **CA-6**

2.1.5. Conversion of CA 2.5 with phenyl isocyanate

CA 2.5 dissolved in pyridine was allowed to react with phenyl isocyanate by using 1:3 molar ratio of reactants (unsubstituted AGU: phenyl isocyanate). Reaction succeeded at room temperature resulting in complete conversion of CA 2.5 to CA-phenylcarbamate (sample **CA-7**). Product obtained was soluble in chloroform and dichloromethane. ^{1}H NMR spectrum of **CA-7** was recorded in CD_2Cl_2 yielded well-resolved AGU showing complete conversion of CA (Fig. 2.7). DS_{Acetyl} 2.26 was calculated from the ratio of spectral integrals of repeating unit and phenyl protons. Spectrum showed three signals of methyl protons assigned to the acetyl moieties of CA 2.5. Aromatic protons of phenylcarbamate appeared at 6.95-7.38 ppm.

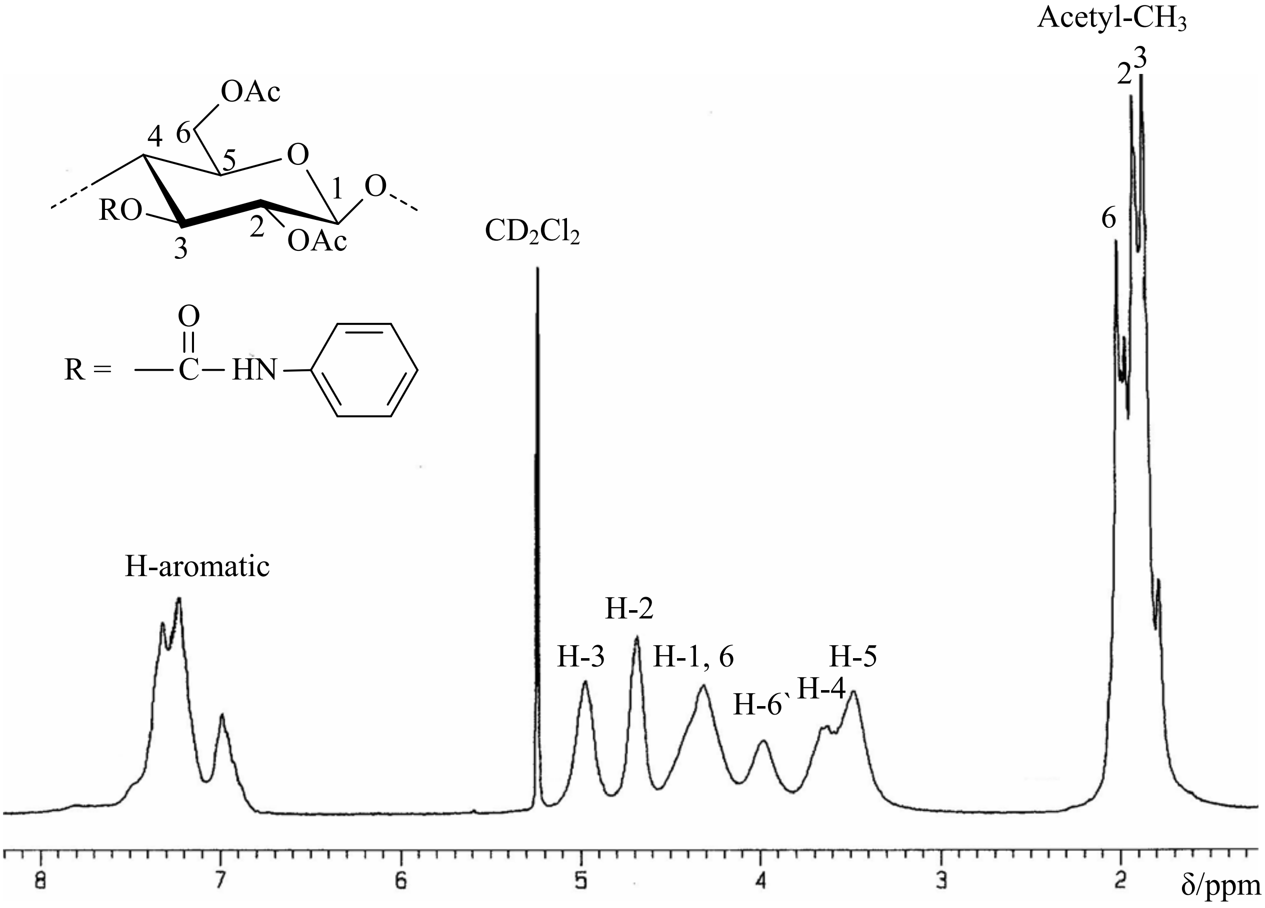


Fig. 2.7: ^{1}H NMR (CD_2Cl_2) spectrum of cellulose acetate phenylcarbamate **CA-7**

Summarising the results one can conclude that uncertainty of DS_{Acetyl} values using different reaction paths mainly depends upon the efficiency of the acylation agents. However, complete substitution of OH groups of CA was possible by acetylation using acetic anhydride and propionylation using propionic anhydride. The results related DS of acetyl functions of CA are more trustable than any other method. These results indicated that CA 2.5 used has DS values of acetyl function 2.32-2.38. Similar results were obtained from ethyl carbanilation, which showed DS of CA 2.43. CA was also possible to analyse using phenyl carbanilation, which showed DS of CA 2.26. Trifluoroacetylation using TFA/CDI was not successful way to substitute completely the free OH groups of CA 2.5. It is also important to note that trifluoroacetylation using TFA/TFA-anhydride has been used for structural analysis of CA. Another valuable method e.g. trimethylsilylation of CA to analyse CA 2.5 has been reported, which showed DS of CA, is 2.28-2.5 (Lee et al., 1995).

Besides structural analysis using ^1H NMR spectroscopy, we calculated DS of the CA 2.5 by using saponification and titration method. DS 2.87 and 2.79 were observed by titration method. The sample of CA 2.5 contains some amount of free acetic acid, which was observed in its ^1H NMR spectrum, goes in titration to shift acetyl value resulted in more DS of the CA. CA 2.5 was re-precipitated from THF into EtOH and then titration was carried out resulted in significant decrease in DS of CA, i.e. 2.70. Re-precipitated sample of CA again re-precipitated by same way and DS of CA 2.63, 2.58 was calculated by titration method.

Concluding, it can be stated that all above-mentioned procedures can be used to analyse CA along with titration method. From all the methods used for structural analysis of CA, acetylation and propionylation using acetic anhydride and propionic anhydride appeared most valuable method to calculate DS of CA by using ^1H NMR spectroscopy.

2.2. New paths for esterification

2.2.1. Esterification of cellulose with fatty acids *in situ* activated with Tos-Cl in *N,N*-dimethylacetamide (DMAc)/LiCl

2.2.1.1. Mechanistic considerations by ^{1}H NMR spectroscopic investigation of the in situ *activation with tosyl chloride*

Acetic acid was reacted with Tos-Cl (1:1 molar ratio) at 80°C for 24 h and ^{1}H NMR spectra of the mixture were recorded. The spectrum indicates the formation of acetic anhydride and acetyl chloride as reactive intermediates (Fig. 2.8). Protons of acetyl chloride appeared at 2.74 ppm while acetic acid anhydride methyl appeared at 2.2 ppm. There are also signals for un-reacted acetic acid and Tos-Cl as indicated in spectrum. *p*-Toluenesulphonic acid (TosOH) formed during the reaction, which is indicated by the presence of signals of aromatic protons as two doublets at 7.22 and 7.61 ppm.

Formation of mixed anhydride of Tos-Cl and acetic acid was supposed to be the reactive intermediate for acetylation of cellulose. However, there is no signal for mixed anhydride of Tos-Cl with acetic acid. The spectrum clearly indicated that the high reactivity of the Tos-Cl as *in situ* activating agent is due to the formation of powerful acylating moieties, i.e. acetyl chloride and acetic anhydride in the reaction medium, which react with cellulose to yield cellulose esters with high efficiency.

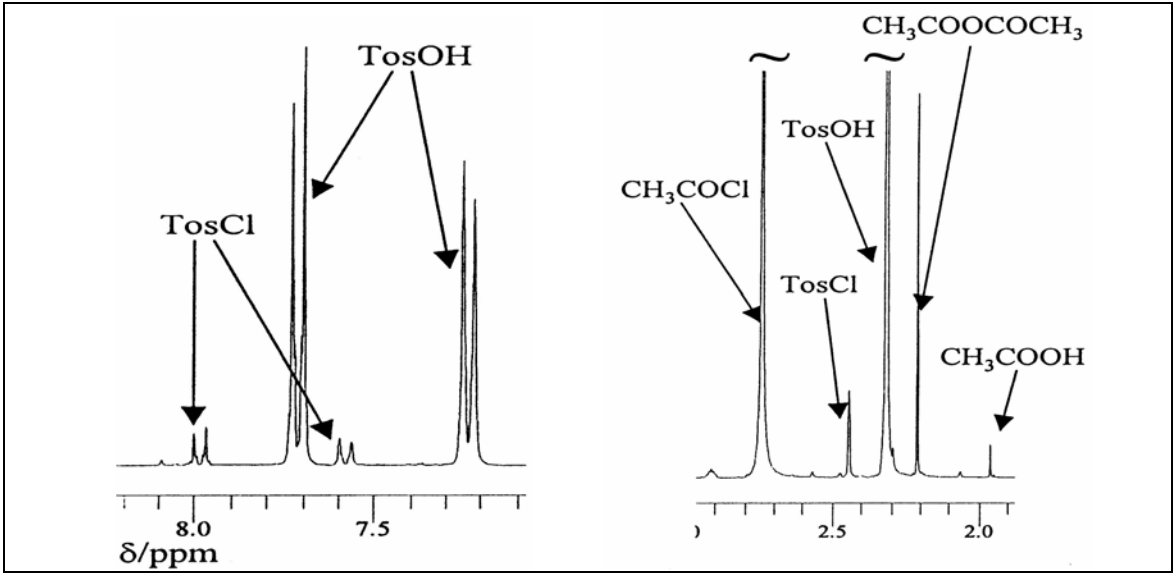


Fig. 2.8: ^{1}H NMR spectrum of the *in situ* activated acetic acid with Tos-Cl

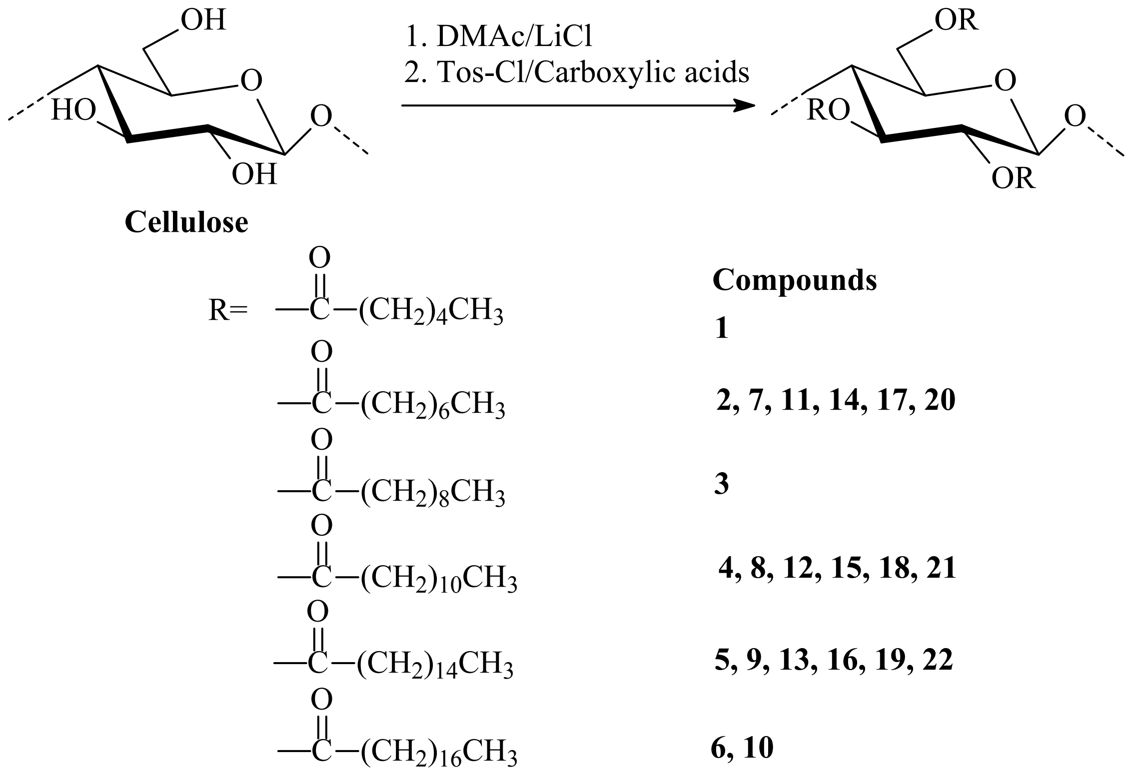
2.2.1.2. Homogeneous acylation of cellulose

An interesting new path for the preparation of cellulose ester is the homogeneous acylation after *in situ* activation of carboxylic acid with Tos-Cl. It was shown that cellulose esters, having alkyl substituents in the range form C12 to C20, could be obtained with almost complete functionalization of the accessible OH groups (Sealey et al., 1996). A variety of different cellulose esters was successfully synthesised via this path, however, without the use of an additional base (Koschella et al., 1997; Heinze and Schaller, 2000).

Considering these results the question arises if the reaction conditions (time, molar ratio of the reagents) and the application of an additional base, e.g. pyridine, influence the degree of substitution, the molecular weight and other structural features of the products. These studies were performed with long chain fatty acids because the efficiency of this particular system for the preparation of the corresponding esters was shown (Heinze and Liebert, 2001).

Thus, cellulose dissolved in DMAc/LiCl was allowed to react with 2 equivalents carboxylic acid (capric-, caprylic-, decanoic-, lauric-, palmitic- and stearic acid) and Tos-Cl without an additional base for 24 h at 80°C (Fig. 2.9). Tos-Cl reacts with carboxylic acids to yield different reactive intermediates, i.e. acid chloride, acid anhydride and mixed anhydride of Tos-Cl and the carboxylic acid, which reacts with the cellulose to yield cellulose esters (Fig. 2.10).

Cellulose esters (polymers **1-6**, Tab. 2.1) were synthesised without base pyridine show two characteristic peaks in FTIR spectra typical for the ester moieties at about 1240 cm^{-1} ($C-O-C_{Ester}$) and about 1750 cm^{-1} ($C=O_{Ester}$). Elemental analysis reveals the absence of sulphur in the samples showing that there is no remarkable introduction of tosylate groups neither covalently bounded nor as impurity.



R=	Compounds
$-C(=O)-(CH_2)_4CH_3$	1
$-C(=O)-(CH_2)_6CH_3$	2, 7, 11, 14, 17, 20
$-C(=O)-(CH_2)_8CH_3$	3
$-C(=O)-(CH_2)_{10}CH_3$	4, 8, 12, 15, 18, 21
$-C(=O)-(CH_2)_{14}CH_3$	5, 9, 13, 16, 19, 22
$-C(=O)-(CH_2)_{16}CH_3$	6, 10

Fig. 2.9: Schematic plot of the conversion of cellulose with carboxylic acid applying *in situ* activation with Tos-Cl

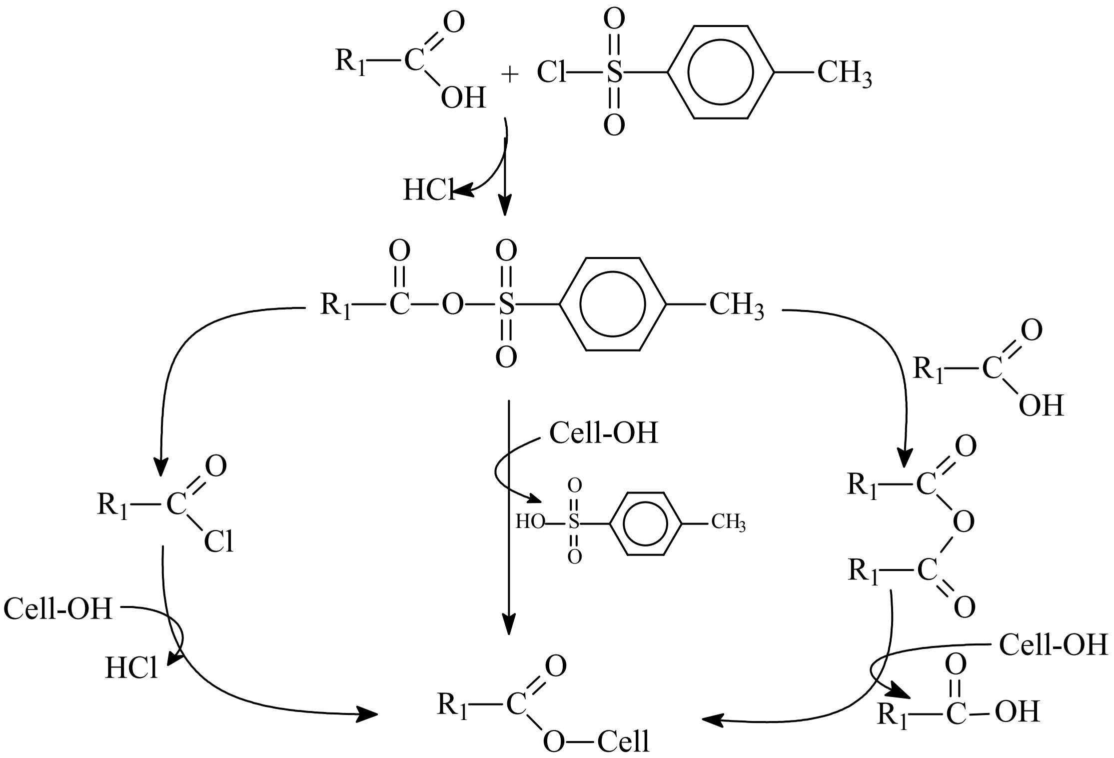


Fig. 2.10: Different reaction routes for cellulose esterification using *in situ* activated carboxylic acid with Tos-Cl

Tab. 2.1: Conditions and results of esterification of cellulose dissolved in DMAc/LiCl mediated with Tos-Cl with different carboxylic acids

	Reaction conditions			Cellulose esters		
Sample	Carboxylic acid	Molar ratio[a]	Time (h)	DS[b]	Yield (g/%)	Solubility
1	Capric	1:2:2:0	24	1.31	2.2/31	DMF, THF
2	Caprylic	1:2:2:0	24	1.40	3.5/42	DMSO, DMF, $CHCl_3$
3	Decanoic	1:2:2:0	24	1.48	4.7/49	DMF, $CHCl_3$, Toluene
4	Lauric	1:2:2:0	24	1.55	8.4/77	Toluene, $CHCl_3$
5	Palmitic	1:2:2:0	24	1.60	10.3/77	Toluene, $CHCl_3$
6	Stearic	1:2:2:0	24	1.76	13.0/84	Toluene, $CHCl_3$
7	Caprylic	1:2:2:4	24	1.76	6.3/66	DMF, $CHCl_3$
8	Lauric	1:2:2:4	24	1.79	9.8/81	$CHCl_3$
9	Palmitic	1:2:2:4	24	1.71	12.8/91	$CHCl_3$
10	Stearic	1:2:2:4	24	1.92	12.8/77	$CHCl_3$
11	Caprylic	1:1:1:0	24	0.63	1.3/22	DMSO, DMF
12	Lauric	1:1:1:0	24	0.36	1.3/23	Pyridine, DMAc/LiCl
13	Palmitic	1:1:1:0	24	0.46	1.7/26	Pyridine, DMAc/LiCl
14	Caprylic	1:4:4:0	24	2.56	10.1/84	Toluene, $CHCl_3$
15	Lauric	1:4:4:0	24	2.56	14.0/90	Toluene, $CHCl_3$
16	Palmitic	1:4:4:0	24	2.54	16.8/89	Toluene, $CHCl_3$
17	Caprylic	1:2:2:0	4	1.27	6.5/82	DMF, $CHCl_3$
18	Lauric	1:2:2:0	4	1.55	9.1/83	$CHCl_3$
19	Palmitic	1:2:2:0	4	1.50	11.1/87	$CHCl_3$
20	Caprylic	1:2:2:0	1	1.25	6.2/79	DMSO, DMF
21	Lauric	1:2:2:0	1	1.36	7.8/77	Insoluble
22	Palmitic	1:2:2:0	1	1.36	8.7/73	$CHCl_3$

[a]) = AGU: carboxylic acid: Tos-Cl: pyridine

[b]) = DS calculated by ^1H NMR spectroscopy after peracetylation

The ^{13}C NMR spectrum of **4** recorded in $CDCl_3$ shows the characteristic signals at $\delta = 173.8$ (CO), 104.0 (C-1), 102.6 (C-1`), 72.3 (C-2), 73.3 (C-3), 82.0 (C-4), 75.1 (C-5), 62.5 (C-6), 13.9 (CH_3) ppm. The signals of the methylene groups of the lauric acid appear in the range of 22.6-34.0 ppm (Fig. 2.11).

The peak for C-6 bearing an ester group appears at δ = 62.5 ppm. The acylated primary OH group exhibits a downfield shift of about 3 ppm compared with the corresponding carbon of the CH_2OH function.

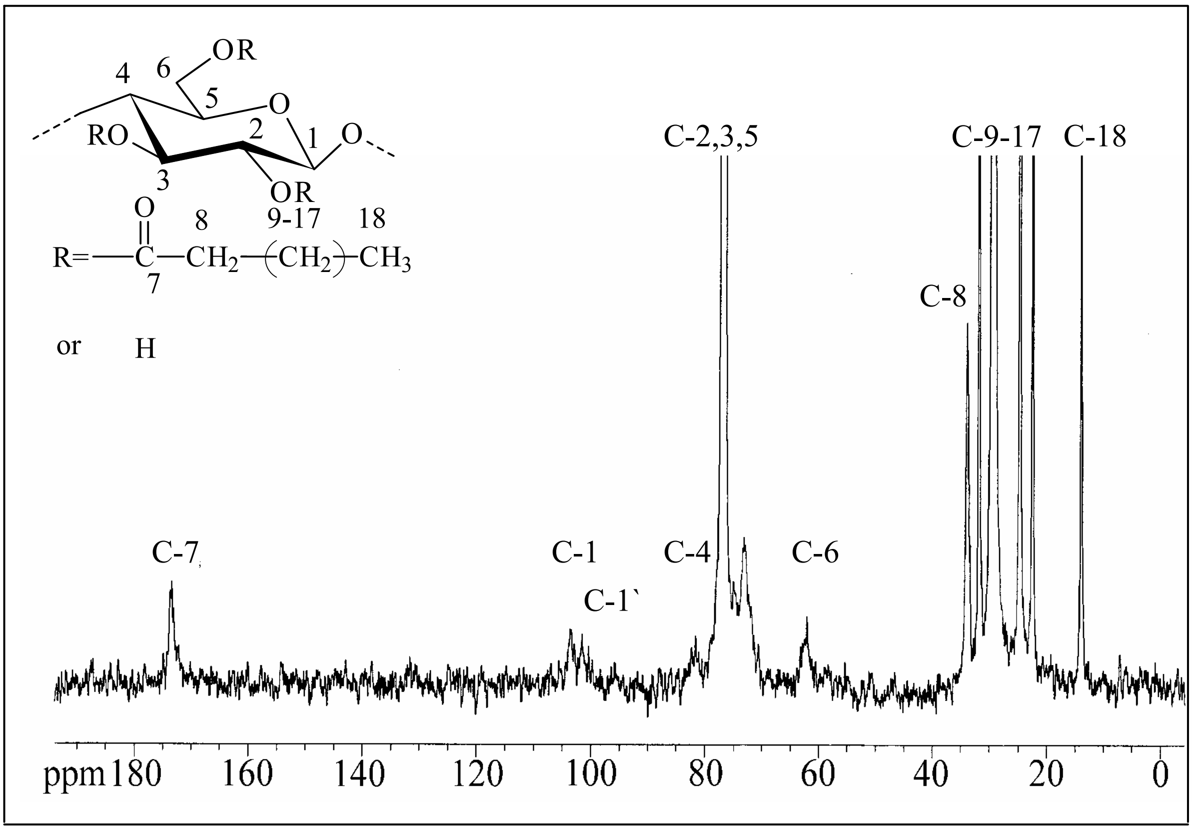


Fig. 2.11: ^{13}C NMR ($CDCl_3$, NS 11,000) spectrum of cellulose laurate **4** (DS = 1.55), index ` means influenced by a functionalization of the neighbour position

To calculate DS, all the cellulose esters **1-22** were completely functionalised using acetic anhydride/pyridine to get peracetylated products **1.1-22.1** (see Tab. 3.2). The peracetylated cellulose esters of fatty acids were readily soluble in $CHCl_3$. A representative ^1H NMR spectrum of cellulose acetate laurate **4.1** (synthesized from sample **4**) recorded in $CDCl_3$ is shown in Fig. 2.12.

The protons of laurate moiety appear at 2.3 (H-8), 1.2-1.6 (H-10-17) and 0.8 (H-18) ppm. The acetate methyl group leads to the signal at 1.9 (H-20) ppm. These results of structure elucidation are in very good agreement with values reported for a cellulose acetate laurate synthesised in the new solvent dimethylsulphoxide (DMSO)/TBAF applying vinyl laurate and acetic anhydride (Ciacco et al., 2003).

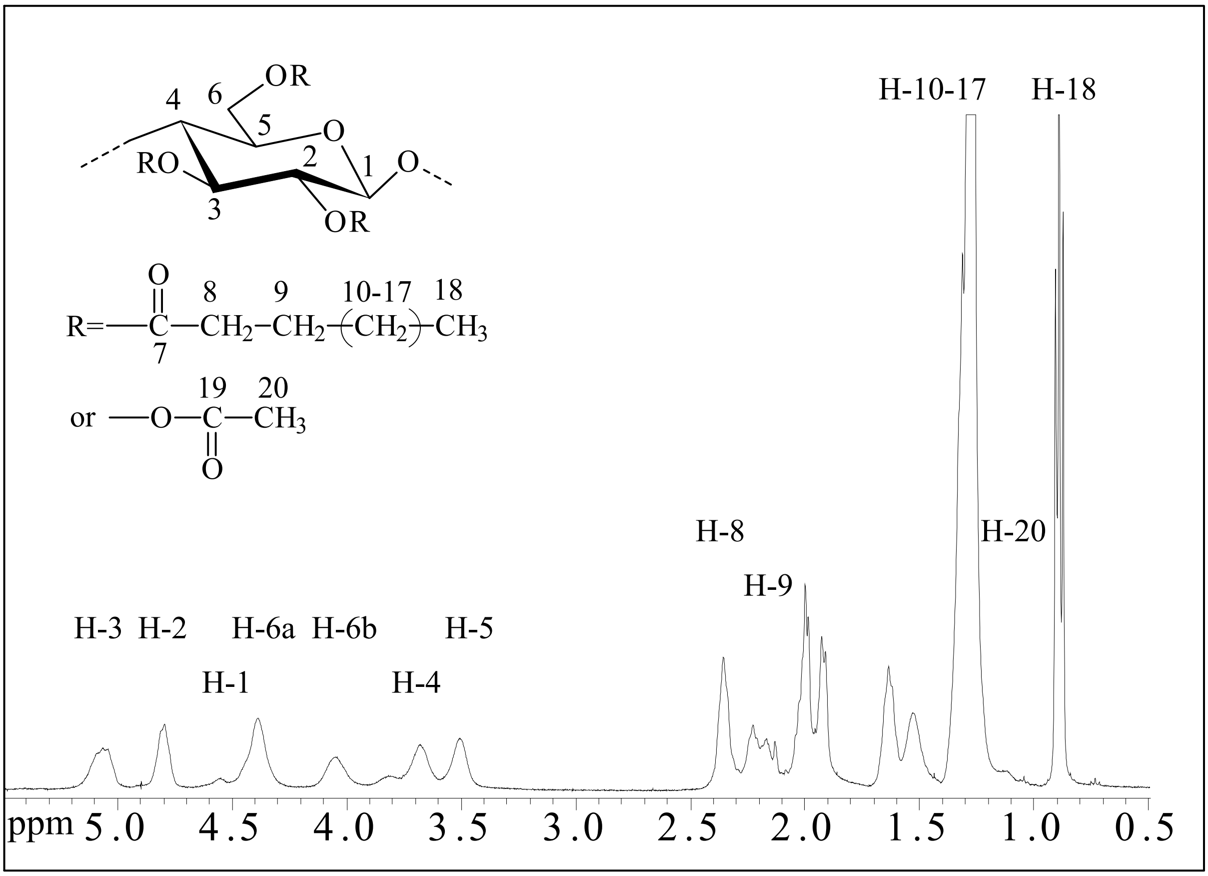


Fig. 2.12: ^{1}H NMR ($CDCl_3$, NS 16) spectrum of cellulose acetate laurate (starting polymer **4**)

It was found that the DS is increased with increasing carbon number of the carboxylic acid. Thus, a DS of 0.6 was found for the cellulose caprate **1** while cellulose caprylate **2** possesses a DS 1.4. Under comparable conditions a cellulose stearate **6** with a DS 2.0 was even accessible.

The cellulose esters possess a different solubility depending on their DS and chain length of the carboxylic acid (Tab. 2.1). In general, cellulose fatty acid esters having DS values higher than 1.4 are soluble in $CHCl_3$ independent of the chain length of carboxylic acid. Polymers with DS values higher than 2.3 are additionally soluble in toluene.

In another series of experiments, the influence of an additional base was investigated. Cellulose was reacted with 2 equivalents of carboxylic acid and Tos-Cl and 4 equivalents pyridine as base. Thus, polymers **7-10** were obtained bearing caprylic- (**7**), lauric- (**8**), palmitic- (**9**) and stearic ester (**10**) functions. It was found that the DS values are higher compared to the samples prepared without base (**1-6**). For instance, a DS of 1.55 was found for the cellulose laurate **4** synthesised without base. The addition of base increases the DS to

1.79 (**8**) (Fig. 2.13). Elemental analysis revealed the absence of sulphur. Therefore, it can be concluded that Tos-Cl acts as activating reagent only. No tosylation occurs.

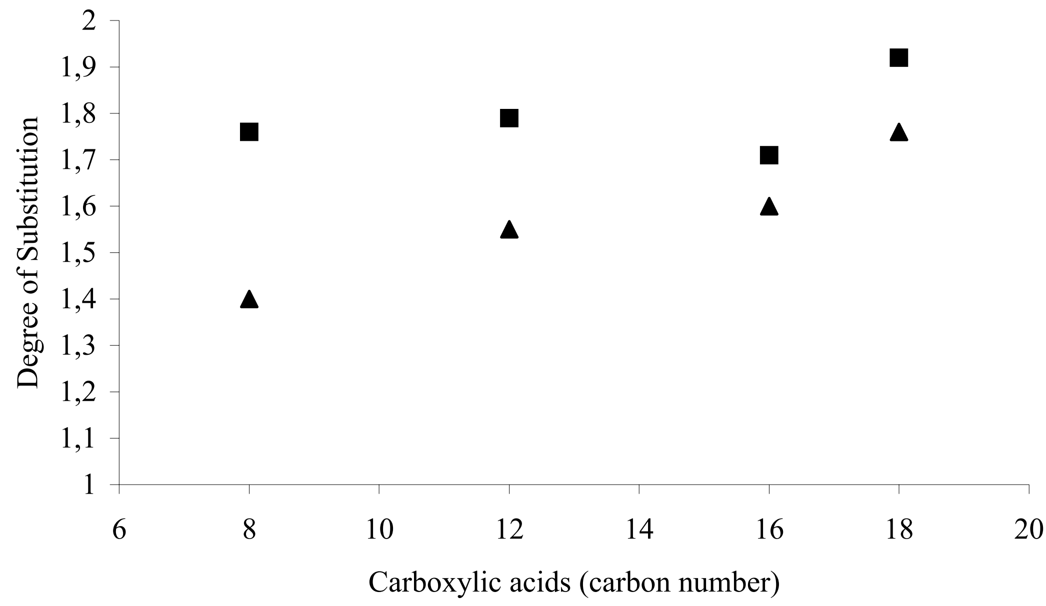


Fig. 2.13: DS of cellulose esters synthesized in DMAc/LiCl using *in situ* activation with the Tos-Cl in dependence on the carboxylic acid and the addition of pyridine (■) and without pyridine (▲)

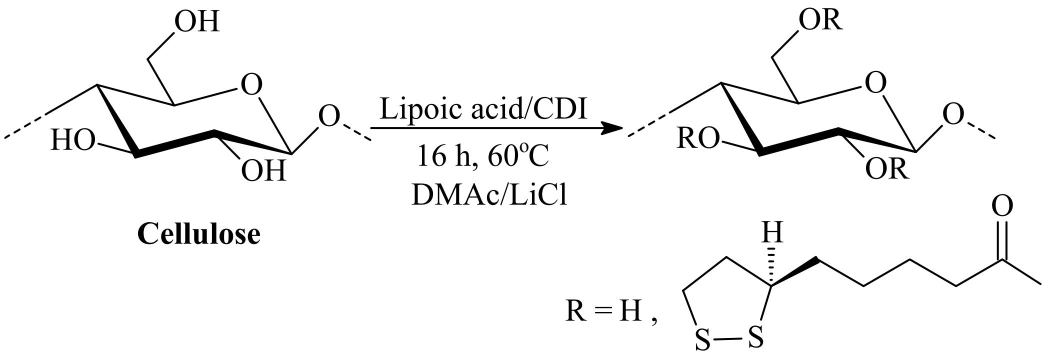
GPC was applied to investigate hydrolytic degradation of the polymer chain during the reaction. Cellulose palmitate **5** synthesised in the absence of base, yielded a polymer with DP 41 whereas cellulose palmitate **9** synthesised in the presence of base, yielded a DP value of 69. Similar results were obtained for cellulose stearate **6** (without base, DP = 45) and **10** (with base, DP = 61). Compared with the DP of the starting cellulose Avicel (DP 280) a fairly drastic degradation occurred in any case.

Thermal decomposition temperatures (*T*d) were obtained from thermogravimetric analysis (TGA) for cellulose caprate (292°C), caprylate (300°C), decanoate (301°C), laurate (302°C), palmitate (306°C) and stearate (318°C). Cellulose esters **1-6** showed the increasing stability with the increase in chain length from C-6 to C-18. Minimum *T*d value of cellulose laurate **4** was 292°C. 318°C was the maximum *T*d value for cellulose stearate **6**. The results of thermogravimetric analysis were comparable with the reported behaviour of long chain fatty acid esters of cellulose (Sealey et al., 1996).

2.2.2. Synthesis and characterization of cellulose α-lipoate prepared by different paths

The esterification of cellulose with α-lipoic acid (thioctic acid) was carried out because this ester moiety contains disulphide function in five membered ring of α-lipoic acid, which can be used for biomineralization after thin layer formation over gold (Bartz et al., 2000). On the other hand, incorporation of α-lipoic acid functions may lead to the product with biological activities due to it's well known antioxidant properties.

For the activation of α-lipoic acid different methods were used, i.e. *in situ* activation with Tos-Cl and CDI. The reactions were carried out homogeneously in DMAc/LiCl. Thus cellulose dissolved in DMAc/LiCl was allowed to react with 3 equivalent α-lipoic acid and Tos-Cl to yield sample **23** (Fig. 2.14).



Compounds	Methods
23	Tos-Cl
24-27	CDI

Fig. 2.14: Schematic plot of the conversion of cellulose with α-lipoic acid *in situ* activated with Tos-Cl and CDI

Reaction carried out at 60°C for 16 h yields organo-insoluble cellulose α-lipoate, however, formation of cellulose α-lipoate was confirmed by FTIR spectroscopy. Important information obtained from FTIR (KBr) spectrum in case of cellulose α-lipoate **23** (Fig. 2.15) was that cyclic ring of α-lipoate moiety remains intact during the reaction as no signal at 2565 cm^{-1} (S-H stretching) appears. Cyclic CH_2 (C-H bending vibrations) appeared as significant signal at 1438 cm^{-1}, which is in the same region of cyclopropane as S-S in the ring of α-lipoate moiety has no effect on the signals of cyclic CH_2. Carbonyl group appeared at 1742 cm^{-1}. Hydroxyl group appeared at 3481cm^{-1}. However, cyclic S-S stretching vibrations usually appear very week in region of 400-500 cm^{-1} does not appear in our spectrum. Spectrum showed successful esterification without disruption of ring.

Similar FTIR spectra were obtained for all of the esters **23-27**. Elemental analyses were carried out for the esters **23-27** (Tab. 2.2) and used to calculate the DS. Proton signals of the tertiary carbon in α-lipoate ring overlap with the cellulose backbone signals, so DS can’t be calculated by ^1H NMR spectroscopy.

While synthesizing cellulose α-lipoate our interest was to prepare organo-soluble cellulose α-lipoate for thin films. For the reason, another method, i.e. *in situ* activation of α-lipoic acid using CDI was studied. Esters **24-27** were synthesized (Tab. 2.2). Samples **25-27** were soluble in DMSO. Perpropionylation of sample **25** was carried out using propionic anhydride/pyridine yielded chloroform soluble sample (**25.1**).

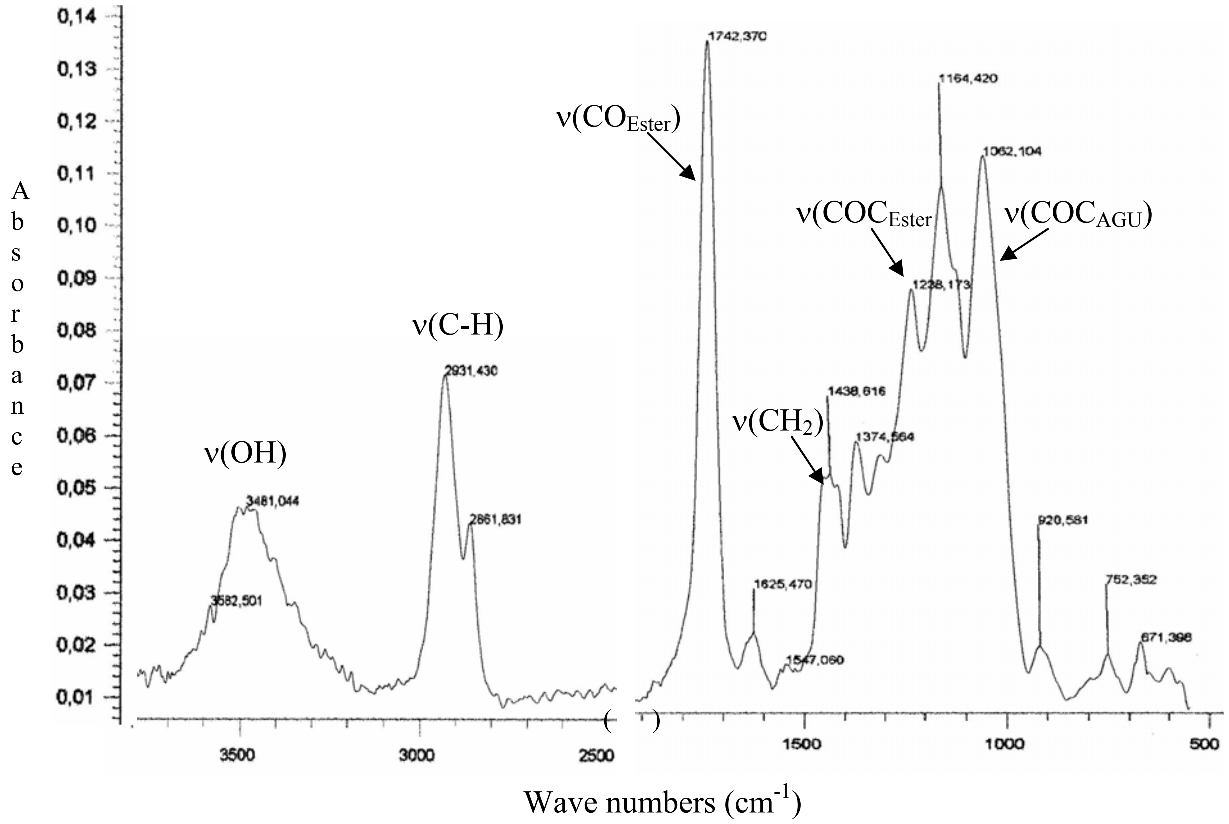


Fig. 2.15: FTIR (KBr) spectrum of cellulose α-lipoate **23** (DS 1.45)

Tab. 2.2: Conditions and results of esterification of cellulose dissolved in DMAc/LiCl with α-lipoic acid *in situ* activated with Tos-Cl **23** and CDI **24-27**

Samples	Molar ratio[a]	Elemental analysis %			DS[b]	Yield (g/%)	Solubility
		C	H	S			
23	1:3:3	48.20	6.04	21.32	1.45	2.50/93	Insoluble
24	1:3:3	45.40	6.78	12.55	0.50	1.33/84	Insoluble
25	1:1.5:1.5	34.81	6.27	5.94	0.18	1.20/99	DMSO
26	1:1:1	42.29	6.64	5.29	0.16	1.15/96	DMSO
27	1:0.5:0.5	41.80	6.81	3.78	0.11	1.10/97	DMSO

[a]) = AGU:α-lipoic acid :Tos-Cl or CDI

[b]) = DS calculated by EA

^{1}H NMR ($CDCl_3$) spectrum of **25** (DS 0.18) (Fig. 2.16) after perpropionylation (**25.1**) showed propionate methyl group (H-15 signal at 0.99, 1.08 and 1.18 ppm) for position 3, 2 and 6, respectively. Signal of H-14 is overlapped with the signal of H-7 at 2.1ppm. Well-resolved AGU appeared at δ = 3.46-5.01 ppm. H-12 and H-13 were appeared at 3.10 ppm as complex signal. Aliphatic chain of α-lipoate moiety H-8-10 appeared at δ = 1.35-1.88 ppm.

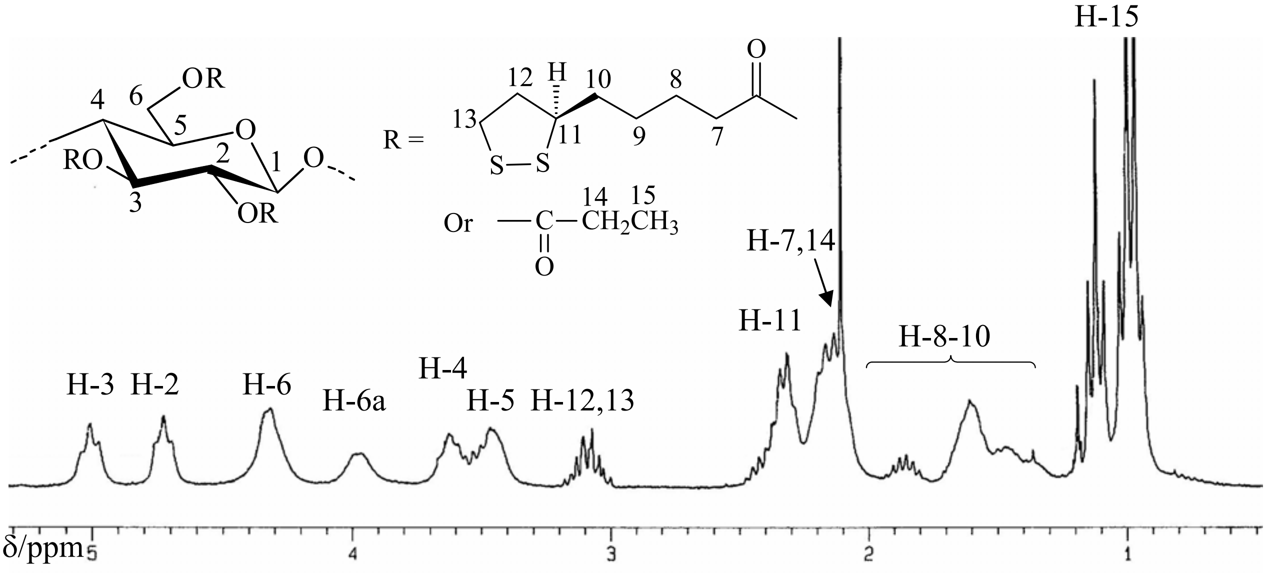


Fig. 2.16: ^{1}H NMR ($CDCl_3$, NS 16) spectrum of cellulose α-lipoate propionate **25.1** (starting polymer **25**)

2.2.2.1. Surface plasmon resonance of thin films of cellulose α-lipoate

Thin films of α-cellulose lipoates (**27**, **26**, **25.1**) were prepared by placing gold slides in a solution of cellulose α-lipoates (2 mmol) in DMSO for 12 h. After washing the surface, binding of the polymer was studied by surface plasmon resonance (SPR). SPR-spectra were recorded against ethanol on bare gold slides. SPR spectra were simulated using a three layers model including the prism, gold and cellulose α-lipoate layers. Refractive indices used were n= 1.7, n=1.84, and n=1.54 respectively for prism, gold and cellulose lipoates.

The angular change in plasmon curve is indicating binding of the cellulose lipoates onto gold surface. The largest shift of the plasmon curve was observed for the thin films of cellulose α-lipoate **27** (DS 0.11) corresponding to angular change of 0.560° comparing bare gold (Fig. 2.17). The simulated film thickness was calculated 49 Å for cellulose α-lipoate **27**. Thin film of cellulose α-lipoate **26** (DS 0.16) yielded shift of the plasmon curve corresponding to angular change of 0.480° comparing bare gold (Fig. 2.18). The simulated film thickness was calculated 29 Å for cellulose α-lipoate **26**.

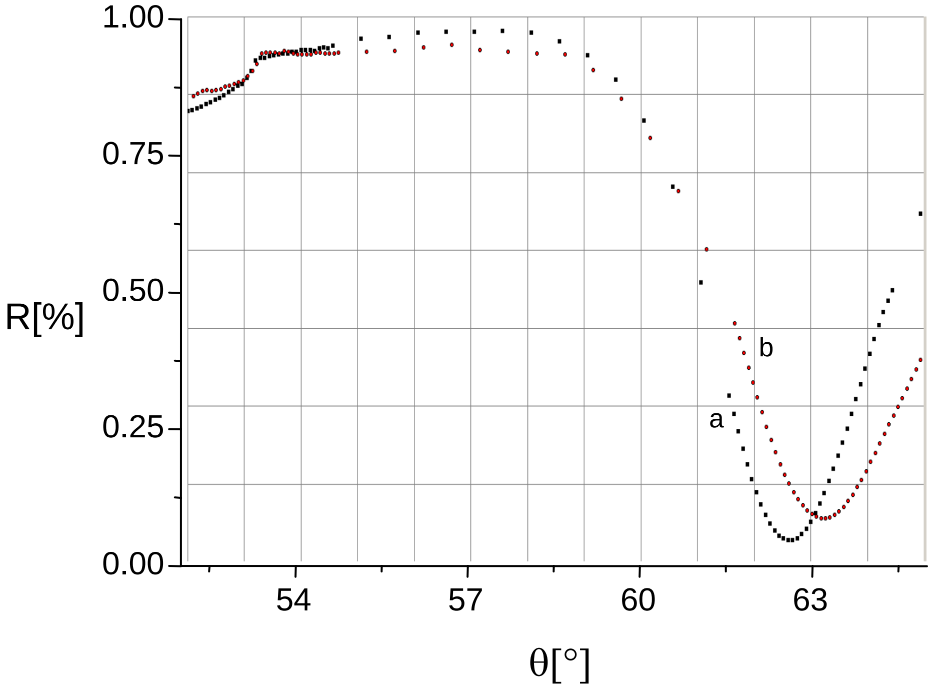


Fig. 2.17: SPR spectrum as function of %age reflectivity (R[%]) vs coupling angle (θ[°]) of the bare gold (a) and coated with cellulose α-lipoate **27** (b)

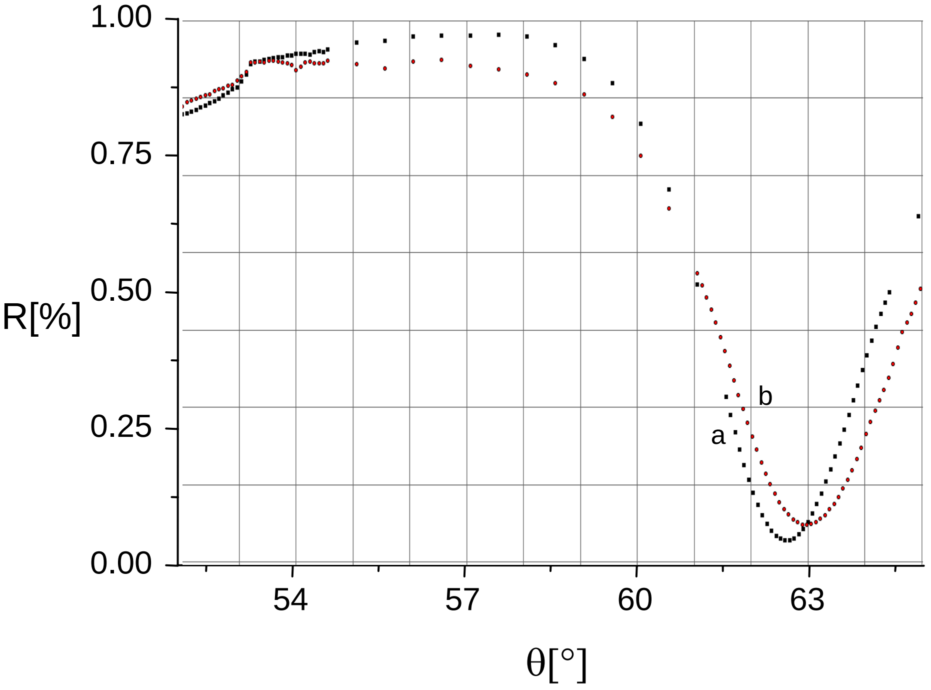


Fig. 2.18: SPR spectrum as function of %age reflectivity (R[%]) vs coupling angle (θ[°]) of the bare gold (a) and coated with cellulose α-lipoate **26** (b)

A perpropionylated cellulose α-lipoate **25.1** (DS 0.18) showed smalest shift of the plasmon curve of 0.160° (Fig. 2.19). The simulated film thickness was calculated 9 Å for cellulose α-lipoate **25.1**.

Consequently, the surface binding can be influenced by changing the DS. Comparably, less substituted cellulose α-lipoate **27** (DS 0.11) yielded more angular change of the plasmon curve due to a more uniform distribution of S-S functions on the cellulose backbone. Low substituted cellulose lipoates showed more surface binding due to less stearic hindrance of the substituted cellulose backbone. Sample **25.1** showed less binding onto gold surface due to rather high DS (0.18) and more stearic hindrance of the substituted cellulose backbone. On the other hand, less stearically hindered backbone of the cellulose ester is usually more flexible in solution, hence higher chain flexibility can results in more binding onto gold surface.

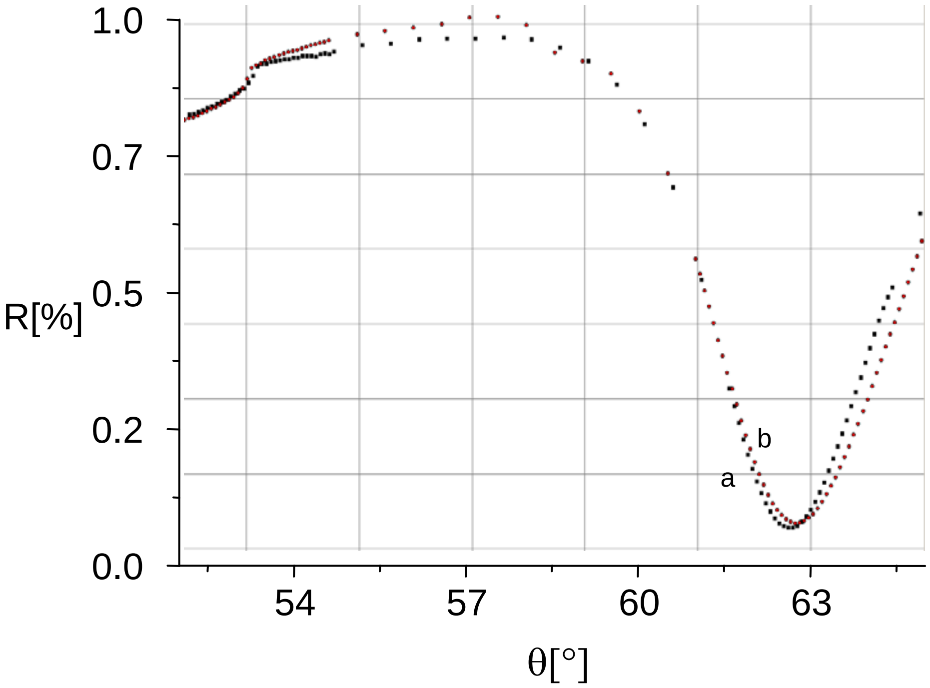


Fig. 2.19: SPR spectrum as function of %age reflectivity (R[%]) vs coupling angle (θ[°]) of the bare gold (a) and coated with cellulose α-lipoate **25.1** (b)

2.2.3. Esterification of cellulose with carboxylic acid *in situ* activated with iminium chloride in DMAc/LiCl

2.2.3.1. Reaction mechanism study of iminium chloride formation

Oxalyl chloride reacts with DMF to form its iminium salt (Stadler, 1978), which reacts with carboxylic acid to form carboxylic acid iminium chloride (see Fig. 2.22). NMR spectroscopy was used to investigate the formation of iminium chloride of carboxylic acid as reactive intermediate. Oxalyl chloride was reacted with DMF and acetic acid. Reaction mixture was kept for 4 h at -20°C then ^{1}H NMR (DMSO-d_6) and ^{13}C NMR (DMSO-d_6) spectra of the mixture of oxalyl chloride, DMF and acetic acid were recorded at room temperature.

^{1}H NMR (DMSO-d_6) spectrum (Fig. 2.20) shows that *N,N*-dimethyl moieties of iminium chloride appeared at δ = 2.65 (H-2) and 2.82 (H-1) ppm while H-3 appears at 7.89 ppm. Acetate methyl appears at 1.82 (H-5) ppm. From the spectrum it is clear that there is no free acetic acid. Formation of acetyl chloride was not observed because as no signals of its methyl protons at 2.66 ppm were observed. Traces of unreacted DMF were also observed in spectrum.

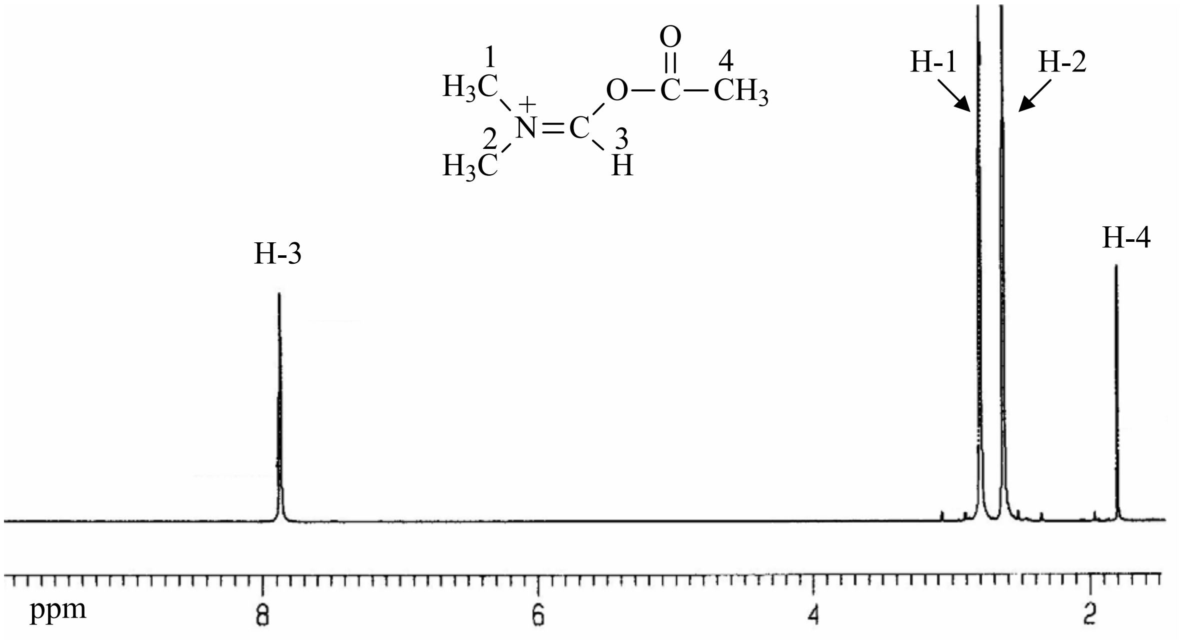


Fig. 2.20: ^{1}H NMR (DMSO-d_6) of acetic acid iminium chloride of acetic acid as reaction intermediate

^{13}C NMR (DMSO-d_6) also proved the formation of iminium chloride (Fig. 2.21). *N,N*-dimethyl moieties of iminium salt appeared at $\delta = 30.30$ (C-2) and 35.42 (C-1) ppm while the tertiary carbon of iminium function appears at 162.04 ppm, which is indicative of the successful formation of iminium chloride. Acetate methyl appeared at 20.03 (C-5) ppm represents the formation of ester function with the iminium chloride moiety. The carbonyl of acetate moiety appears at 171.54 (C-4). There is no signal for free acetic acid methyl and its carbonyl that normally appears at about $\delta = 21.7$ and 176.9 ppm respectively. Formation of acetyl chloride is not observed because of the absence of signals of both corresponding methyl carbon at 33.6 ppm and carbonyl at 170.4 ppm in ^{13}C NMR spectrum. ^{1}H NMR and ^{13}C NMR spectra prove the formation of iminium chloride as reactive intermediate at above mentioned reaction conditions.

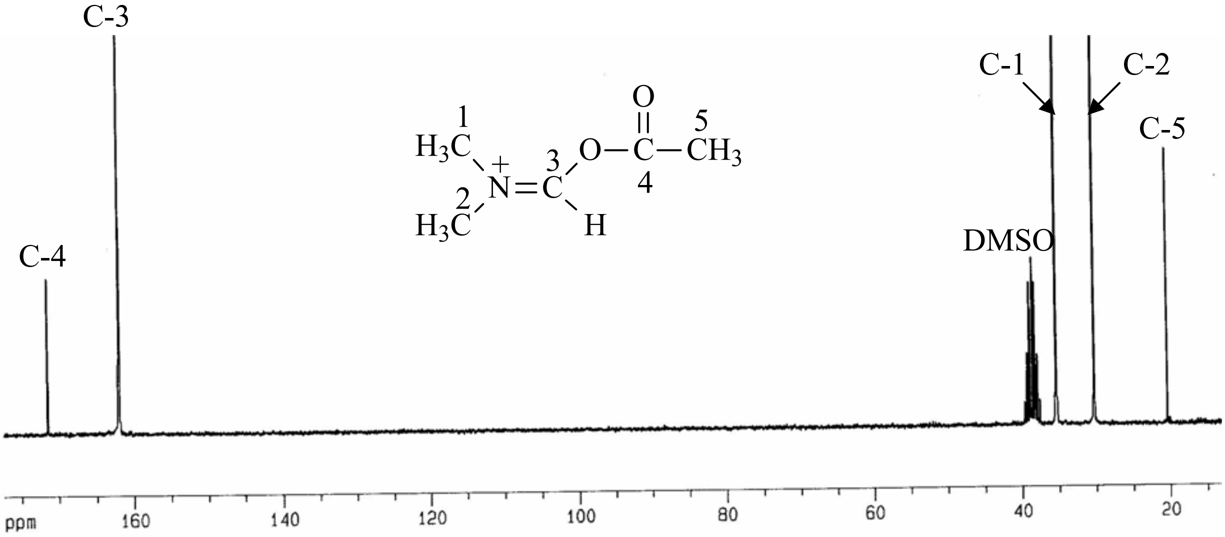


Fig. 2.21: ^{13}C NMR (DMSO-d_6) spectrum (NS 820) of iminium chloride of acetic acid as reaction intermediate

2.2.3.2. Homogenous acylation of cellulose

To study the potential of the method for the esterification of cellulose carboxylic acids with different substructures were reacted via the iminium chlorides, i.e. acetic acid, the long chain aliphatic acids stearic acid and palmitic acid, the aromatic acid 4-nitrobenzoic acid and adamantane 1-carboxylic acid as bulky alicyclic acid. The formation of the iminium chloride and the conversion with the acid were carried out as “one pot reaction”, i.e. DMF was cooled to –20°C, oxalyl chloride was added very carefully and after the gas-formation had stopped the carboxylic acid was added. The conversion succeeds with quantitative yield at this temperature. The complex is stable and no side reactions like the formation of HCl and acid chloride were observed as can be confirmed by NMR spectroscopic studies.

This mixture was added to a solution of 1.0 g cellulose in DMAc/LiCl. The reaction was carried out at 60°C for 16 h. During the reaction at 60°C for 16 h gelation occurred. The purification is rather easy because most of the products are gaseous and during the last step DMF formed again (Fig. 2.22). Moreover, the cellulose ester floats on the reaction mixture if stirring is stopped at the end of the conversion and can be isolated in very good yields simply by filtration and by washing with ethanol. A summary of reaction conditions and results is given in Tab. 2.3.

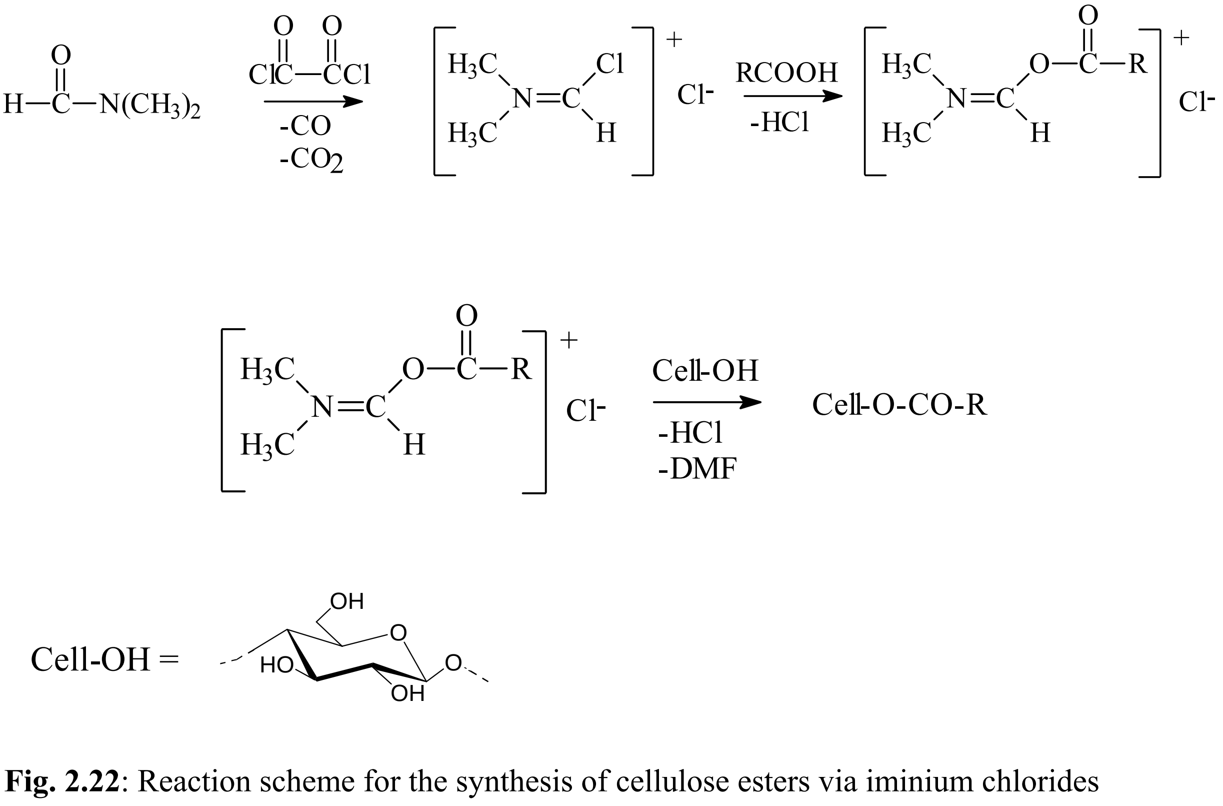


Fig. 2.22: Reaction scheme for the synthesis of cellulose esters via iminium chlorides

The method is suitable for the preparation of different type of cellulose esters. It is especially efficient for the esterification with aliphatic and alicyclic carboxylic acids (samples **28-36**). DS values as high as 1.89 (sample **32**) was achieved by this reaction path. Increasing DS values can be observed for molar ratios of carboxylic acid to anhydroglucose unit (AGU) of up to 1:3. If the ratio is in the range of 1:6 the solution became highly viscous or even a thick gel during the reaction resulting in decreasing DS values (sample **36, 40**). In case of the acetylation of cellulose highly functionalized esters were obtained (analyzed by FTIR spectroscopy) but these derivatives were insoluble in common organic solvents. This insolubility was also observed for cellulose acetates prepared with acetyl chloride (without base) and via CDI (Heinze et al., 2003). It is still a matter of discussion if this behaviour is due to cross-linking or an unconventional superstructure caused by the complete acetylation of the primary hydroxyl function and/or an uneven distribution of the acetyl groups within the polymer chains.

Tab. 2.3: Conditions and results of esterification of cellulose dissolved in DMAc/LiCl mediated with oxalyl chloride with different carboxylic acids

Sample	Carboxylic acid	Mole ratio[a]	DS[b]	Yield (g/%)	Solubility
28	Stearic acid	1:1:1	0.15	0.89/70	DMAc/LiCl, DMSO/LiCl
29	Stearic acid	1:2:2	0.21	0.52/38	DMAc/LiCl, DMSO/LiCl
30	Stearic acid	1:3:3	0.63	1.80/86	DMAc/LiCl, DMSO/LiCl
31	Stearic acid	1:5:5	1.84		THF, $CHCl_3$
32	Palmitic acid	1:6:6	1.89		DMSO, DMAc, THF
33	Adamantane 1-carboxylic acid	1:1:1	0.47	0.63/43	DMSO/LiCl
34	Adamantane 1-carboxylic acid	1:2:2	0.52	0.48/31	DMAc/LiCl, DMSO/LiCl
35	Adamantane 1-carboxylic acid	1:3:3	1.20	0.80/34	DMAc, DMSO, DMF
36	Adamantane 1-carboxylic acid	1:6:6	0.66	1.14/66	DMSO
37	4-nitrobenzoic acid	1:1:1	0.30	0.60/46	DMAc/LiCl, DMSO/LiCl
38	4-nitrobenzoic acid	1:2:2	0.52	0.65/42	DMSO
39	4-nitrobenzoic acid	1:3:3	0.94	1.02/42	DMSO
40	4-nitrobenzoic acid	1:6:6	0.66	1.14/68	DMAc/LiCl, DMSO/LiCl

[a]) = AGU: carboxylic acid: oxalyl chloride

[b]) = DS calculated by ^{1}H NMR spectroscopy after peracetylation (**28-32**) and perpropionylation (**33-40**)

The cellulose esters were characterized by means of FTIR spectroscopy, EA, ^{1}H NMR and ^{13}C NMR spectroscopy as well as ^{1}H NMR spectroscopy after peracetylation (**28-32**) or perpropionylation (**33-40**). The FTIR (KBr) spectra showed typical absorption for the polysaccharide backbone (3620, 2920 and 1140 cm^{-1}) and signals for the carbonyl function of the ester moiety at 1745-1760 cm^{-1}. Elemental analysis of all cellulose esters was carried out and found in agreement with DS values calculated by ^{1}H NMR spectroscopy after peracylation (see Tab. 3.3). DS values for cellulose stearate **28-30** were calculated by EA, are 0.16, 0.18 and 0.44, respectively, are comparable with the DS values 0.15, 0.21and 0.63 obtained by ^{1}H NMR spectroscopy after peracetylation. DS values (0.25 and 0.60) were calculated by EA for cellulose adamantoate **33** and **36** are comparable with the DS values 0.47 and 0.66 obtained by ^{1}H NMR spectroscopy after perpropionylation. A representative ^{1}H NMR (CDCl$_3$) spectrum of cellulose 4-nitrobenzoate **39** after perpropionylation (sample **39.1**) is shown in Fig. 2.23. The spectrum showed signals of anhydroglucose unit at δ= 3.46-5.04 (H-1-6) ppm and for the aromatic protons of nitro-benzoate moiety at 7.79-8.31 (H-7, 8) ppm. The propionate ethyl group leads to the signal at 2.10 (H-9) ppm and propionate methyl group appears at 0.99 (H-10) ppm.

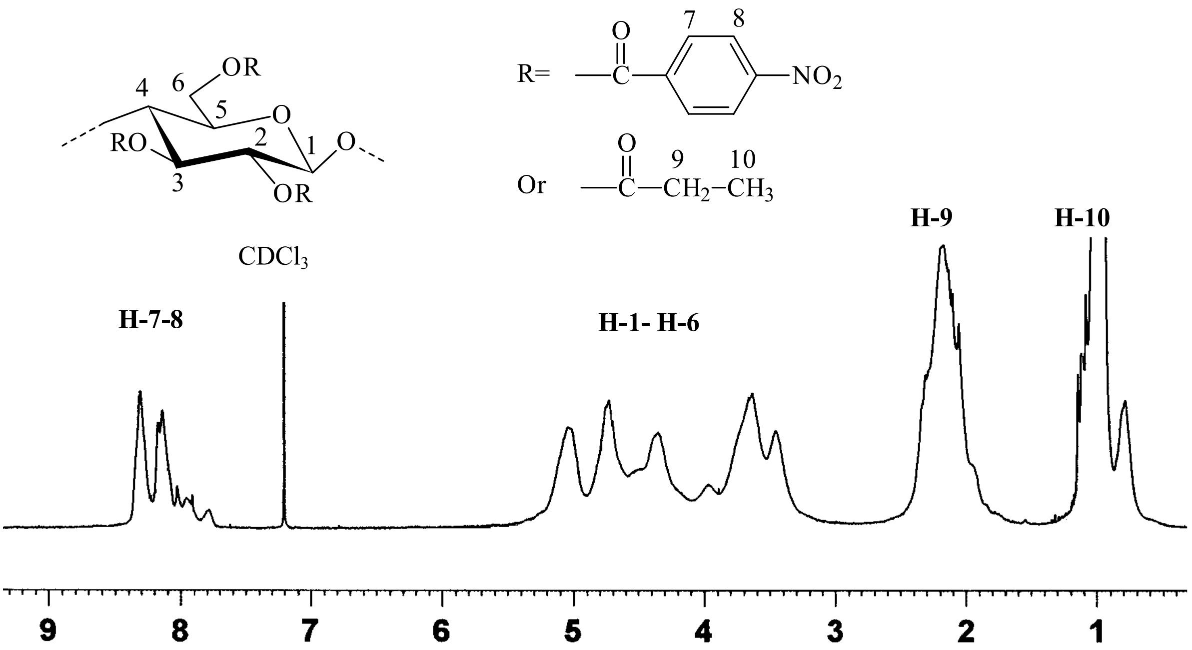


Fig. 2.23: ^{1}H NMR spectrum (CDCl$_3$) of cellulose 4-nitrobenzoate propionate **39.1** (DS 0.94, starting polymer **39**) after perpropionylation

DS values given in Tab. 2.3 were calculated from ^{1}H NMR spectra after acylation of the remaining hydroxyl groups according to our own standard method (Heinze et al., 2000). EA confirmed the DS calculated by ^{1}H NMR spectroscopy. It needs to be mentioned that the samples contained up to 2-3 % chlorine. The nature of the impurity has not been clarified yet. Up to now there is no evidence for the existence of deoxochloro functions.

Gel permeation chromatography (GPC) was applied to obtain information about the degradation of the cellulose backbone during the conversion. DP values of 240 for cellulose adamentate (DS 1.20, **35**), 280 for cellulose 4-nitrobenzoate (DS 0.52, **38**) and 250 for cellulose stearate (DS 1.84, **31**) were obtained if Avicel® with DP 280 was the starting polymer. Thus, esterification via iminium chlorides are much milder compared to conversion via *in situ* activation with tosyl chloride or functionalization with the acid chlorides (Heinze et al., 2003).

Summarizing it can be stated that the method is a very efficient tool for the synthesis of pure aliphatic, aromatic and bulky cellulose esters with high DS values and minimum degradation. It seems to be especially valuable for the synthesis of aliphatic esters. Products with DS values as high as 1.89 can be prepared by “one pot synthesis” at very moderate reaction conditions yielding polymers soluble in THF. The DS values are comparable to values obtained for the conversion via activation with tosyl chloride (Heinze et al., 2003). However, the important advantage of the iminuim chloride method is a diminished degradation of the polymer chain during the conversion generally in the range of less than 15% and a very easy work up procedure. Comparable results can be obtained for adamantoyl cellulose (samples **33-36**).

Comparison with samples prepared with the acetyl chloride (maximum DS 1.94) and via activation with tosyl chloride (maximum DS 1.75) and *N,N'*-carbonyldiimidazole (maximum DS 1.42) show the activation with iminium chloride gave the product of the highest DP. Consequently, this type of esterification combines a high efficiency with very mild reaction conditions. Thus, it might be possible to exploit this path for the synthesis of sophisticated or sensitive esters, e.g. with unsaturated or chiral moieties. The method is a rather inexpensive and could be applied at large scale.

2.2.4. Synthesis of cellulose esters with carboxylic acids *in situ* activated with CDI in DMSO/TBAF

2.2.4.1. Mechanism of imidazolide formation

CDI reacts with carboxylic acid to give imidazolide of the carboxylic acid and CO_2 (Gräbner *et al.*, 2002; Staab, 1962). For studying the reaction mechanism propionic acid was allowed to react with CDI in DMSO as solvent. Reaction mixture was kept under stirring for 24 h at room temperature to facile complete conversion of acid to it's imidazolide. ^{1}H NMR (DMSO-d_6) spectrum of propionic acid imidazolide is shown in Fig. 2.24). The spectrum showed that propyl moiety of imidazolide appears as triplet at δ 0.95 (H-1) and quartet at δ 2.18 (H-2) ppm. Aromatic protons from imidazolide moiety appear as three separate signals at δ 7.84 (H-3), 6.17 (H-4), and 7.08 (H-5) ppm. Spectrum is showing complete conversion of propionic acid to it's imidazolide, which is evident from the absence of signals of unreacted propionic acid in the spectrum.

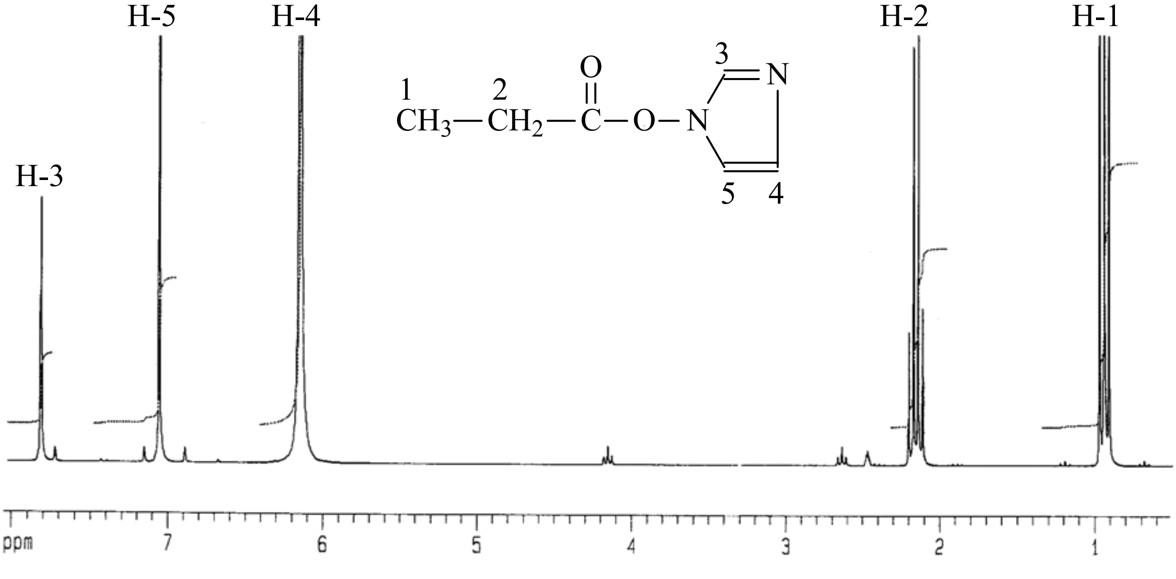


Fig. 2.24: ^{1}H NMR (DMSO-d_6, NS 16) spectrum of propionic acid imidazolide as reaction intermediate

^{13}C NMR spectrum was recorded in DMSO-d_6 (Fig. 2.25) showed signals of propyl moiety at $\delta = 8.99$ (C-1) and 27.16 (C-2) ppm. Aromatic carbons appear at $\delta = 134.94$ (C-3) and 121.13

(C-4, 5) ppm. The imidazolide carbonyl appears at 175.91 (C-6) ppm, however, carbonyl of CDI appears normally at 193 ppm, which is absent, proved it's successful conversion to imidazolide. Free imidazole signals overlap with signals of carboxylic acid imidazolide. There are no signals for the unreacted propionic acid carbons that would appear at about δ = 9.6 (CH_3), 28.5 (CH_2), and 180.4 (CO) ppm. ^{1}H NMR and ^{13}C NMR spectroscopic studies proved the formation of imidazolide of propionic acid as reactive intermediate.

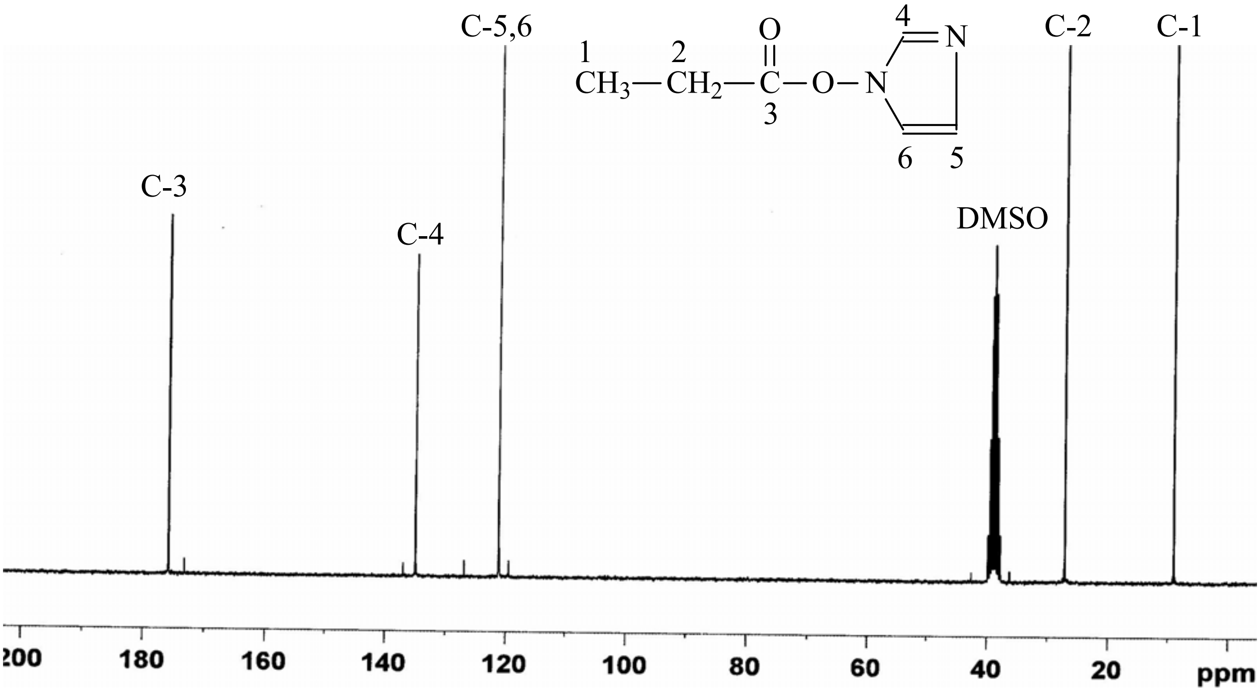


Fig. 2.25: ^{13}C NMR (DMSO-d_6, NS 820) spectrum of propionic acid imidazolide as reaction intermediate

2.2.4.2. Acylation of cellulose via imidazolides

Avicel dissolved in DMSO/TBAF was allowed to react with imidazolides of different carboxylic acids, i.e. acetic acid, propionic acid, lauric acid, stearic acid, adamantane 1-carboxylic acid, α-lipoic acid and 2-furan carboxylic acid and cellulose esters **41-50** were synthesized. Imidazolides were prepared by *in situ* conversion of the carboxylic acids with CDI in DMSO at room temperature. The only by products liberated from the reaction are imidazole and CO_2 (Fig. 2.26).

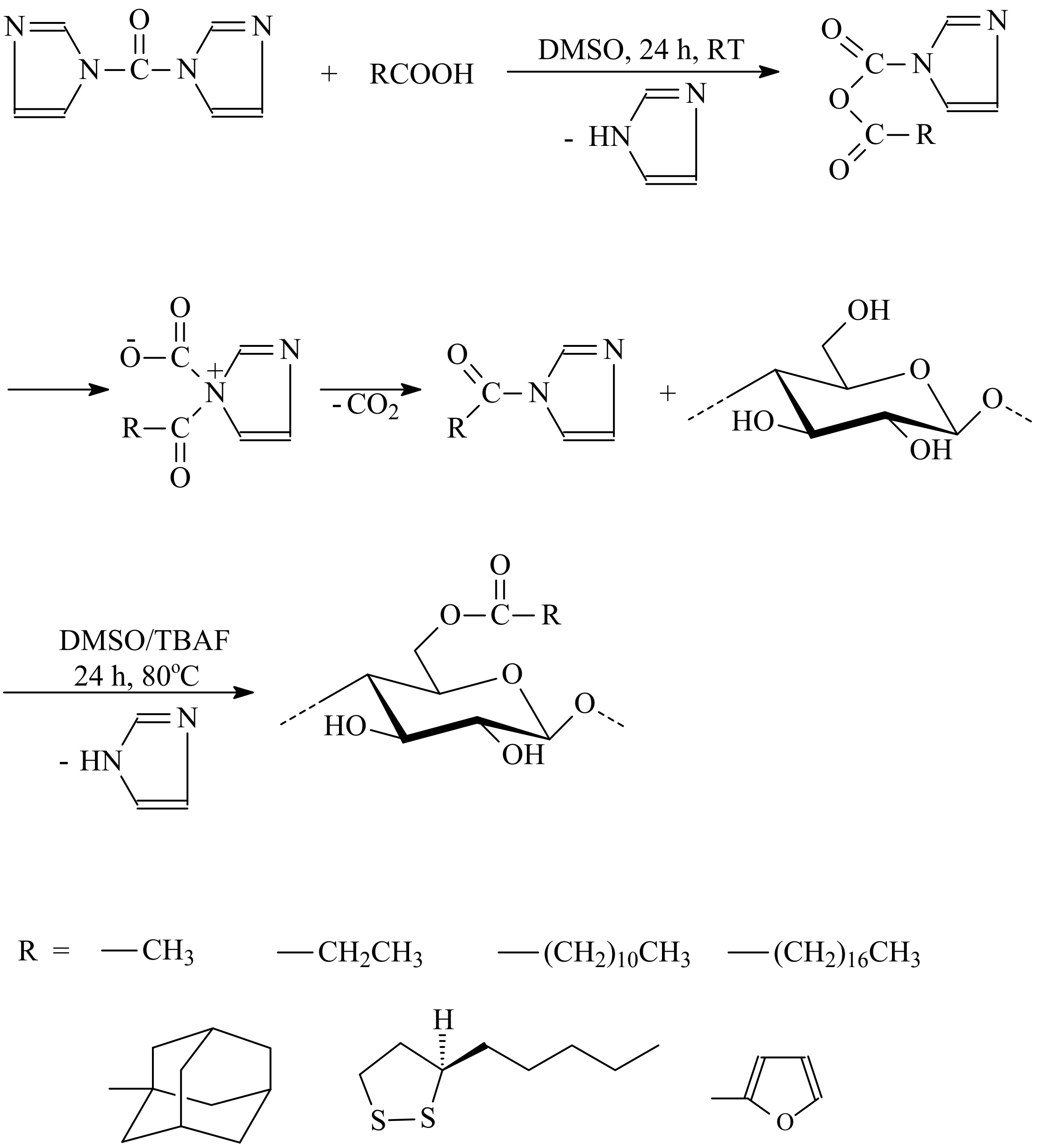


Fig. 2.26: Scheme for cellulose esterification with carboxylic acids applying *in situ* activation with CDI

The acylation of cellulose was simply carried out by adding the solution of imidazolide to the solution of cellulose at increasing temperature. Pure products were obtained by precipitation in EtOH and by filtration. Reaction conditions and results are summarized in Tab. 2.4. By this reaction path, DS values as high as 2.23 (sample **43**) can be achieved. The cellulose esters were characterized by means of FTIR spectroscopy, elemental analysis, ^{1}H NMR and ^{13}C NMR spectroscopy as well as ^{1}H NMR spectroscopy after peracylation. The small chain aliphatic esters 41 & 42 show DS values up to 0.50 & 1.02 if molar ratios (AGU:reagent) 1:3 were applied.

Tab. 2.4: Conditions and results of esterification of cellulose dissolved in DMSO/TBAF mediated with CDI with different carboxylic acids

Samples	Carboxylic acid	Mole ratio[a]	DS[b]	Yield (g/%)	Solubility
41	Acetic acid[c]	1:3:3	0.51	0.92/78	DMSO, DMAc
42	Propionic acid[c]	1:3:3	1.02	0.48/33	DMSO, DMAc
43	Lauric acid	1:3:3	2.23	2.20/62	DMSO
44	Stearic acid	1:1:1	0.05	0.82/75	DMSO
45	Stearic acid	1:2:2	0.47	1.43/79	DMSO
46	Stearic acid	1:3:3	1.35	2.89/86	DMSO
47	α-Lipoic acid [d]	1:3:3	1.22	2.20/86	DMSO
48	Adamantane 1-carboxylic acid	1:2:2	0.50	0.83/53	DMAc/LiCl
49	Adamantane 1-carboxylic acid	1:3:3	0.68	0.87/50	DMSO, DMAc
50	2-Furan carboxylic acid	1:3:3	1.91	1.40/61	DMSO, DMAc

[a]) = AGU: carboxylic acid: CDI

[b]) = DS calculated by ^{1}H NMR spectroscopy after peracetylation (**42-46**) and perpropionylation (**41, 47-50**)

[c]) = Synthesized at 100°C

[d]) = DS calculated by EA

The DS values reached are lower compared to esters prepared using anhydrides, or prepared by imidazolide formation using DMAc/LiCl as solvent. One reason of low reactivity of short chain acid imidazolides (e.g. acetic acid) is due to it’s high reactivity towards hydrolysis, while long chain aliphatic acids imidazolides were less affected by the water of TBAF in results higher DS values were obtained, i.e. DS 2.23 (sample **43**).

An interesting new product synthesized via this path is the α-lipoic acid ester (sample **47** DS 1.22), which is soluble in DMSO. The ester **47** is being studied for thin film formation over gold and will also be studied for bio-mineralization using different metal oxides, e.g. TiO_2.

Cellulose furoate (sample **50**) will be studied for membrane formation to filter proteins, so it is valuable aspect of synthesis.

Elemental Analysis

Elemental analyses were carried out for the esters **41-50**, DS (1.22) of cellulose α-lipoate **47** was possible to calculate only with the help of EA using sulfur as reference atom. However, DS 0.37 and 0.50 calculated by EA for **45** and **50**, which are comparable with DS 0.47 and 0.68 of same esters respectively, while calculated with ^{1}H NMR spectroscopy after peracylation.

Analysis of FTIR spectra

The FTIR (KBr) spectra showed typical absorption for the polysaccharide backbone, signals for the carbonyl function of the ester moiety and aromatic absorptions. A typical FTIR (KBr) spectrum of cellulose furoate **50** prepared from a homogeneous solution with a DS of 1.91 is shown in Fig. 2.27. The spectrum displayed hydroxyl group absorption at 3493 cm^{-1}, aromatic C-H absorption at 3142 cm^{-1}, carbonyl group appeared at 1728 cm^{-1} and aromatic furan ring absorption at 1580 cm^{-1}. Spectrum showed the success of reaction due to carbonyl, aromatic, and ester absorptions appeared, which are in good agreement with the values available in literature for cellulose furoate (Hon *et al.*, 2001, 2001a). In case of cellulose α-lipoate **47**, spectrum displayed hydroxyl group absorption at 3466 cm^{-1}. Important information obtained from FTIR spectrum in case of cellulose α-lipoate **47** is that, cyclic ring of α-lipoate moiety remains intact during the reaction as; the signal at 2565 cm^{-1} (S-H stretching) does not appear. Cyclic CH_2 (C-H bending vibrations) appeared as significant signal at 1440 cm^{-1}. Carbonyl group appeared at 1738 cm^{-1}. However, cyclic S-S stretching vibrations usually appear very week in region of 400-500 cm^{-1} do not appear in our spectra.

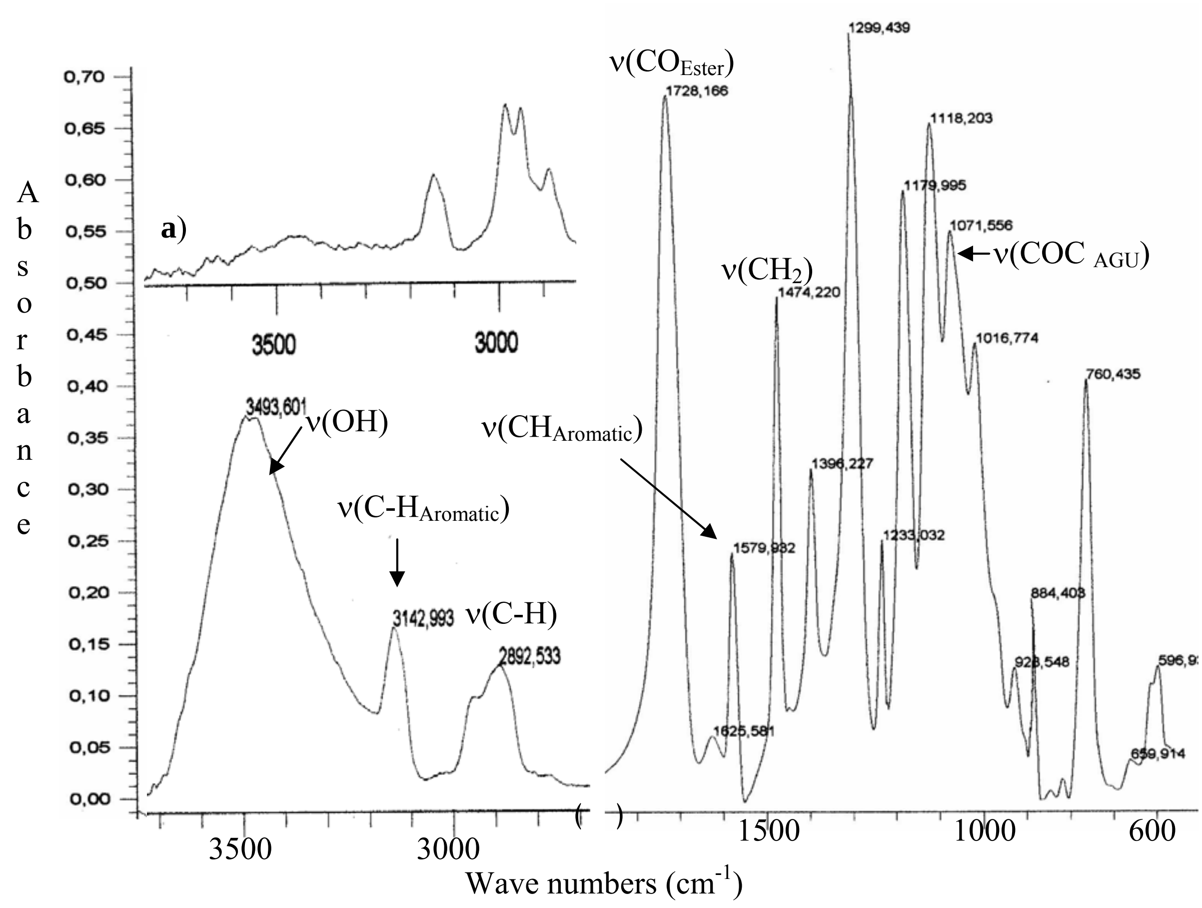


Fig. 2.27: FTIR (KBr) spectra of cellulose furoate **50**, **a**) FTIR spectra (OH region) after complete perpropionylation of **50**

^{1}H NMR spectroscopic characterization

After peracylation, the samples were soluble in chloroform. ^{1}H NMR ($CDCl_3$) spectrum of perpropionylated cellulose furoate **50.1** (Starting polymer **50**) is shown in Fig. 2.28. Protons of propionate moiety appeared at δ 2.04 (CH_2) and 0.77 and 0.93 (CH_3-2, 3-propionate) ppm. DS was calculated from the ratio of the spectral integrals of the protons of the repeating unit (δ 3.63-5.00 ppm) and the methyl protons of the propionate (δ 0.77, 0.93 ppm). DS can be calculated in same manner from protons of the furan ring. Aromatic furan ring protons were detectable at δ 6.50, 7.20 and 7.56 ppm showed that the unsaturated system is not destroyed during the conversion. FTIR spectrum has already indicated aromatic ring absorption. All results for cellulose furoate are comparable with the values available in literature (Hon *et al.*, 2001a).

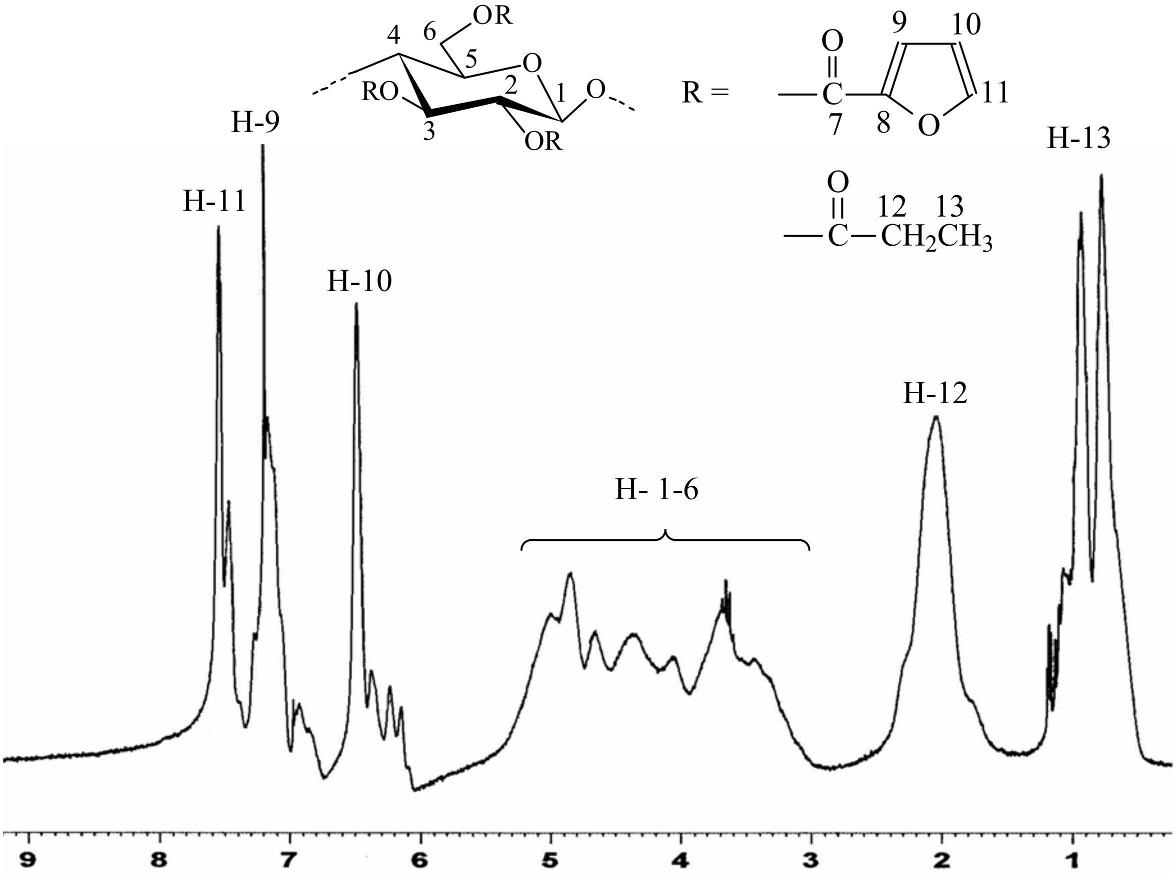


Fig. 2.28: ^{1}H NMR ($CDCl_3$, NS 16) of cellulose furoate propionate **50.1** (starting polymer **50**)

Another important product was cellulose adamantoate (DS 0.68). For the determination of the DS value, cellulose adamantoate **49** was perpropionylated yielding the mixed esters **49.1** without any unmodified hydroxyl functions, which is proved by FTIR spectroscopy (no ν(OH) signals appear). Assignment of the signals in ^{1}H NMR spectrum to the corresponding protons is given in Fig. 2.29. DS can readily calculated from the spectral integrals of the protons of the repeating unit (δ = 3.45-5.05 ppm) and the methyl protons of the propionate (δ = 0.96, 1.11 ppm). DS can be calculated in same manner from the protons of the adamantoyl unit (δ = 1.67-2.31 ppm).

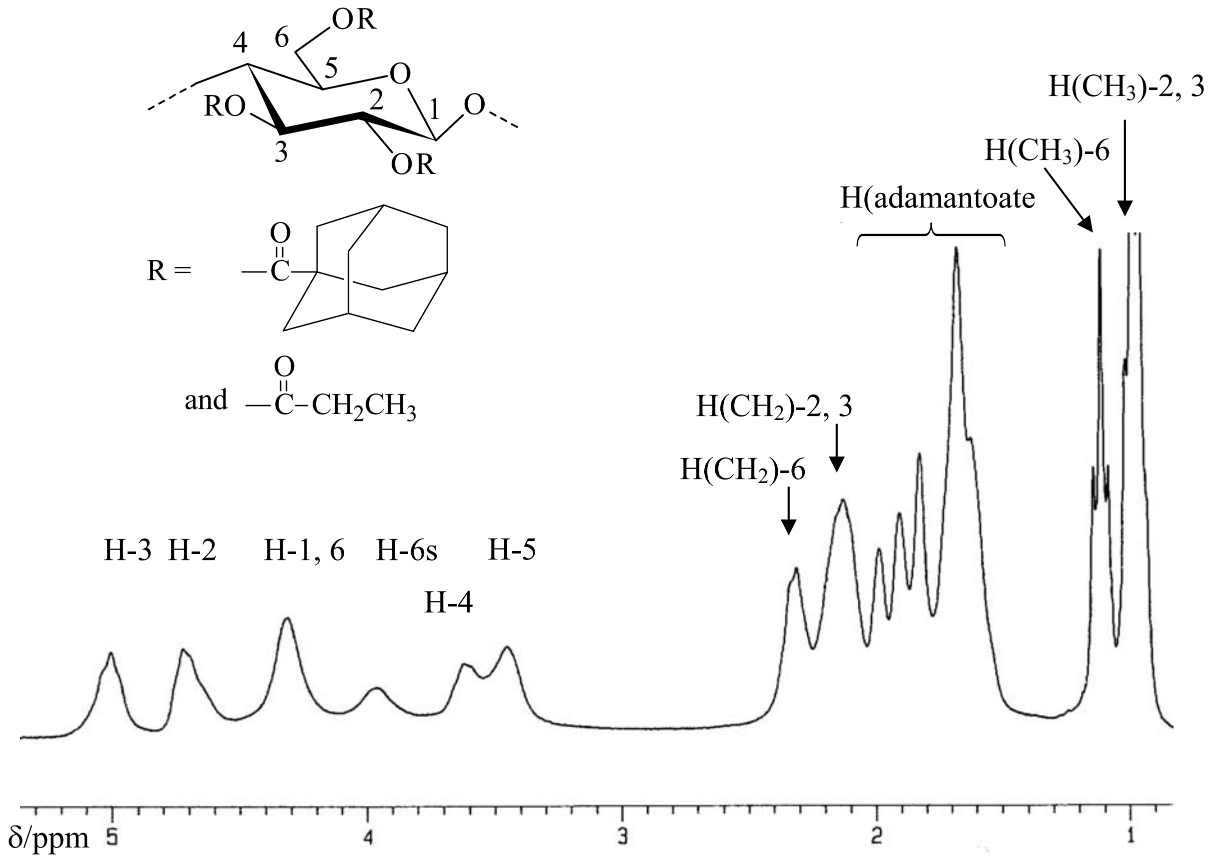


Fig. 2.29: ^{1}H NMR ($CDCl_3$, NS 16) spectrum of adamantoyl cellulose after perpropionylation (starting polymer **49**)

^{13}C NMR spectroscopic characterization

Cellulose furoate **50** were synthesised with DS 1.91 yielded well-resolved ^{13}C NMR spectrum recorded in DMSO-*d*$_6$ (Fig. 2.30). Resonances assigned to the carbon atoms of the furan ester moieties are visible at δ = 143.4 (C-8), 118.8 (C-9), 112.1 (C-10) and 147.6 (C-11) ppm. Carbonyl of furoate moiety appeared at δ157.3 ppm. The signals of the carbon atoms of the modified AGU are detectable in the region δ = 102.9 to 63.1 ppm. The peak for C-6 influenced by esterification in O-6 appears at δ = 63.1 ppm (C-6), i.e. it exhibits a downfield shift of about 3 ppm compared with the corresponding carbon of pure cellulose, which indicates preferred substitution at primary hydroxyl while in case of cellulose adamantoate primary hydroxyl is not completely substituted is indicative of uniform distribution of adamantoyl moiety over the cellulose backbone. The signals at δ = 73.9 and 75.7 ppm result from the carbon atoms at position 2 and 3 are well resolved. The signals of C-1 appear at δ =

103.7 ppm. In addition, the spectrum shows a signal at 99.8 ppm, which corresponds to C-1 adjacent to a C-2 atom bearing a furan moiety.

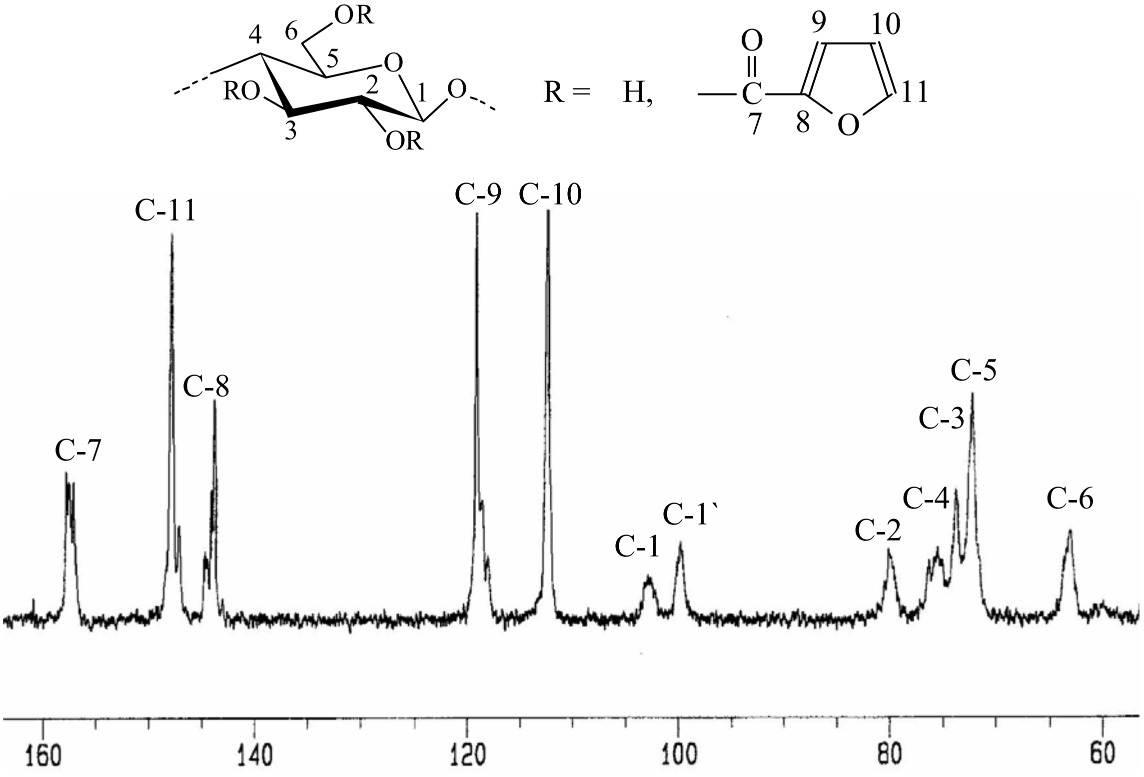


Fig. 2.30: ^{13}C NMR (DMSO-d_6, NS 68,000) spectrum of cellulose furanoate **50**

Gel permeation chromatographic studies

GPC studies revealed chromatograms with a bimodal distribution. The low-molecular weight fraction was assigned to polymers dissolved in a molecular-dispersed manner. The high-molecular weight fraction represents aggregated polymer chains. The signals of the two fractions overlap. Only for the cellulose acetate (sample **41**) and the adamantoyl cellulose (sample **49**) proper evaluation was possible. The depolymerization is rather small. Product **49** possesses a DP of 228 and sample **41** DP 187. The starting cellulose Avicel® had a DP of 280.

2.2.5. Synthesis and characterization of pullulan abietates

Keeping in view the importance of pullulan derivatives in food, cosmetics and pharmaceutics and electronic fields (LeDuy et al., 1989; Bruneel et al., 1993, 1993a, 1994), amphiphilic pullulan abietate (**51-56**) were synthesised using *in situ* activation with Tos-Cl (Fig. 2.31). Results are summarized in Tab. 2.5.

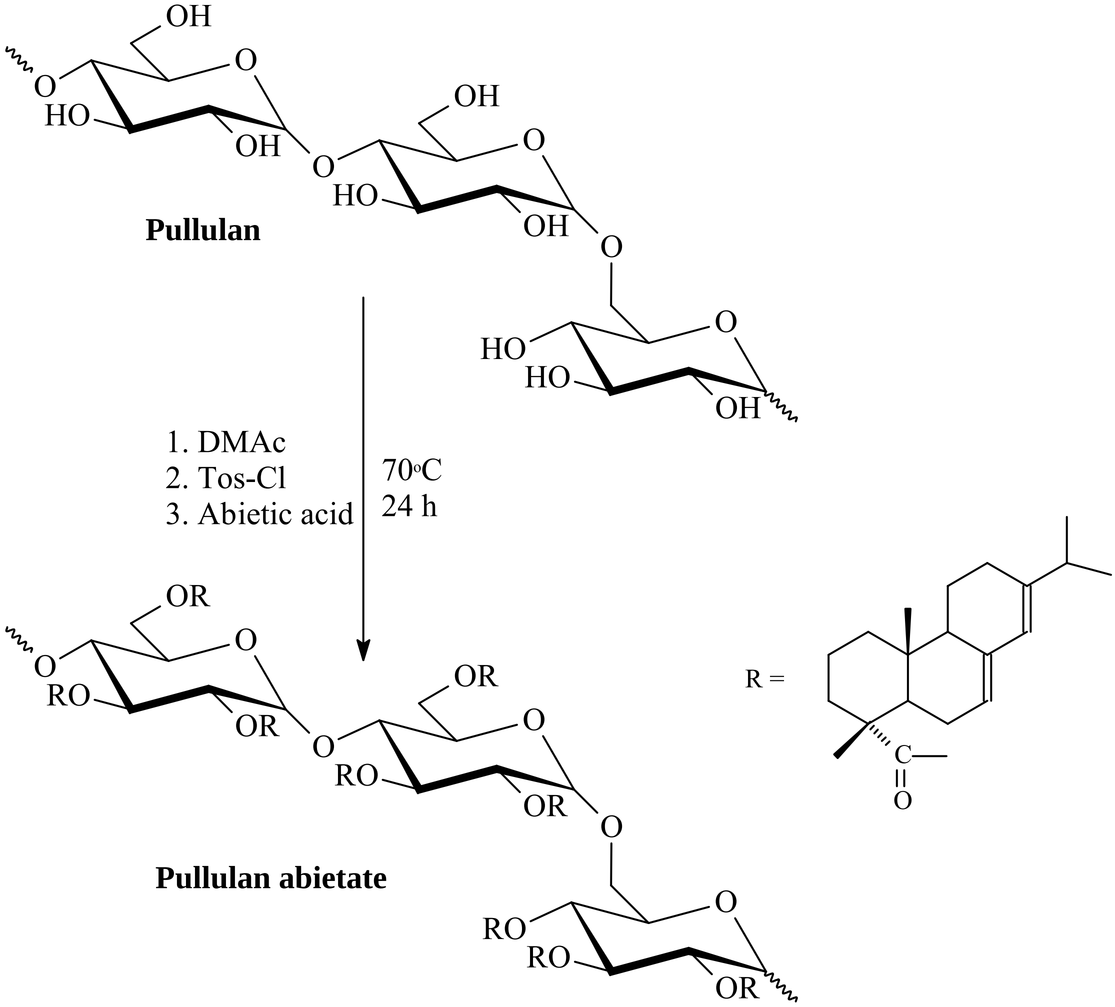


Fig. 2.31: Schematic plot of the conversion of pullulan with abietic acid applying *in situ* activation with Tos-Cl

Pullulan dissolved in DMAc was allowed to react with abietic acid and Tos-Cl at elevated temperature. To get water-soluble product, pullulan esters were synthesized with different molar ratios and with or without pyridine. Compound **51** with low DS of 0.04 appeared water-soluble and was used for self-assembly and to adsorb onto regenerated cellulose surface over gold. Sample **52** and **53** were insoluble in water, however, samples **54-58** are water-soluble. All pullulan abietates **51-58** are readily soluble in DMSO and DMAc. No generalization can be made for water solubility and DS of the abietate but we can conclude that very low substitution of pendant group, i.e. abietic acid, can convert the hydrophilic polymer into amphiphilic polymer.

Tab. 2.5: Conditions and results of the reactions of pullulan dissolved in DMAc with abietic acid after *in situ* activation with Tos-Cl (method A), or CDI (method B), or oxalyl chloride (method C)

Method	Sample	Molar ratio[a]	DS[b]	Yield (g)	Solubility
A	**51**	1:0.5:0.5:1	0.04	4.4/81	Water, DMSO, DMAc, DMF
A	**52**	1:1:1:2	0.06	5.0/90	DMSO, DMAc, DMF
A	**53**	1:1.5:1.5:3	0.25	6.3/88	DMSO, DMAc, DMF, THF
A	**54**	1:0.75:0.75:0	0.10	0.8/68	Water, DMSO, DMAc
A	**55**	1:0.50:0.50:0	0.08	0.7/61	Water, DMSO, DMAc
A	**56**	1:0.25:0.25:0	0.12	0.8/66	Water, DMSO, DMAc
B	**57**	1:1:1:0	0.10	0.7/60	Water, DMSO, DMAc
C	**58**	1:1:1:0	0.07	0.7/62	Water, DMSO, DMAc

[a]) = AGU:Tos-Cl: abietic acid: pyridine

[b]) = DS calculated by titration method (samples **51-52**) or by EA (samples **53-58**)

Success of reaction was established by FTIR spectroscopy, EA and NMR spectroscopic studies. Products of low DS are hard to characterize, however, FTIR (KBr) spectrum of **52** shows two characteristic peaks typical for the ester moieties at about 1246 cm^{-1} (C-O-C$_{Ester}$) and about 1724 cm^{-1} (CO$_{Ester}$) (see Fig. 2.32). Comparable FTIR spectra were obtained for other pullulan abietates.

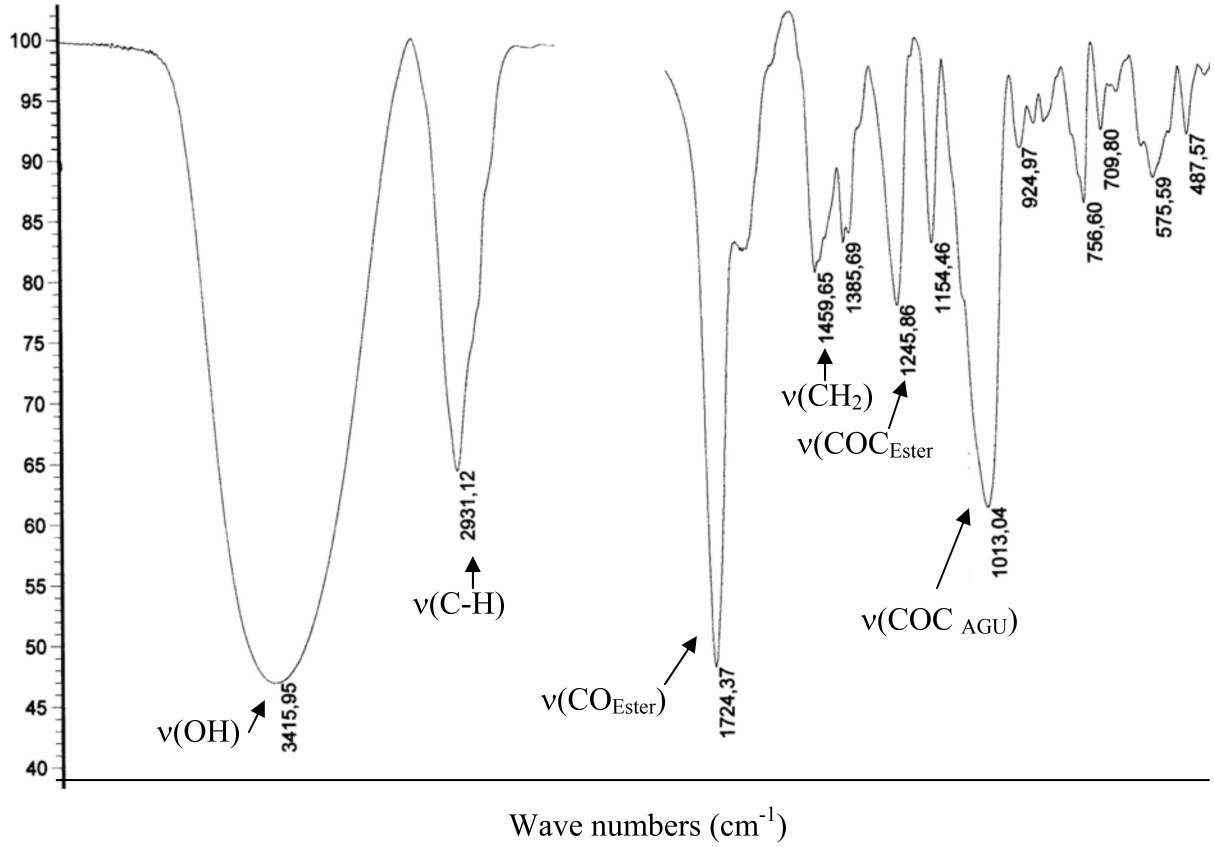


Fig. 2.32: FTIR (KBr, %transmittance) spectrum of pullulan abietate (sample **52**)

Elemental analysis reveals the absence of sulphur in the samples showing that there is no introduction of tosylate groups neither covalently bounded nor as impurity. DS of abietates was also possible to calculate with the help of elemental analysis, (see Tab. 2.6, **53-58**). Samples **54-58** contain 0.1-0.5 % nitrogen traces, which result from DMAc is no significant impurity. However, sample **58** prepared from oxalyl chloride (C-method) contains 2.05% Cl, which was also observed for the cellulose esters synthesised by this method (see section 2.2.3).

It was found that DS of pullulan abietate obtained could be controlled by the molar ratio of abietic acid to AGU. Product of higher DS 0.25 was synthesised by using 1: 1.5 mole ratio of abietic acid to AGU (sample **53**), however, lower DS 0.06 was obtained for sample **51**, which was synthesised using 1/0.5/0.5 molar ratio of AGU/Tos-Cl/abietic acid.

Tab. 2.6: Elemental analyses of pullulan abietates (**51-58**) synthesised by different paths

Compounds	**51**	**52**	**53**	**54**	**55**	**56**	**57**	**58**
DS[a]	0.06	0.17	0.25	0.10	0.08	0.12	0.10	0.07
% C	44.67	51.79	55.91	41.70	42.16	42.42	43.38	40.83
% H	6.87	7.26	7.42	7.29	7.14	7.40	7.20	7.14

[a]) = DS calculated by EA

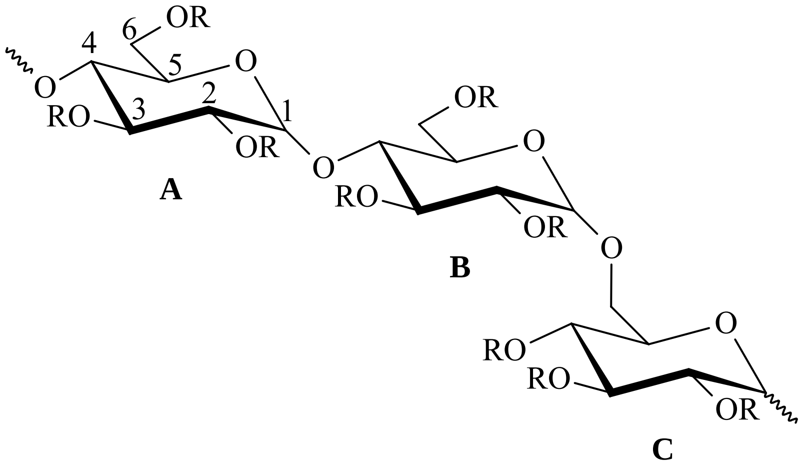


Fig. 2.33: Structure of maltotriose repeating unit of pullulan

Typical ^{1}H NMR (DMSO-d_6) spectrum of **52** (DS 0.06) showed AGU signals of all protons of maltotriose units (see for structure Fig. 2.33) together in the range of 3.26-5.5 ppm (AGU-H), protons of abietate moiety appearing in the range of 0.73-2.75 ppm. Protons at unsaturated carbons of abietate rings appeared at 5.71(H-20) and 5.31 (H-13, overlapped with AGU) ppm (see Fig. 2.34).

A typical ^{13}C NMR spectrum of **52** recorded in DMSO-d_6 shows the characteristic carbonyl peak at δ = 177.4, 170.5and 168.1 ppm, which is valuable information for the success of reaction (Fig. 2.35). Three signals indicate the substitution of abietic acid at C-1 of sugars A and B in maltotriose unit of pullulan. Well resolved AGU shows signals at δ = 101.7, 99.3, 96.0 (C-$1_{A,\ B,\ C}$), 80.4 (C-$4_{A,\ B}$), 60.9-73.8 (C-2, 3, 4_C, 5, 6). Unsaturated carbons showed signals at δ = 120.8 (C-13), 122.9 (C-20), 134.9 (C-14) and 144.7 (C-19) ppm. ^{1}H NMR and ^{13}C NMR spectra of pullulan abietate are comparable for AGU (maltotriose region) with the ^{1}H NMR and ^{13}C NMR spectra of pullulan peracetate (Tezuka, 1998).

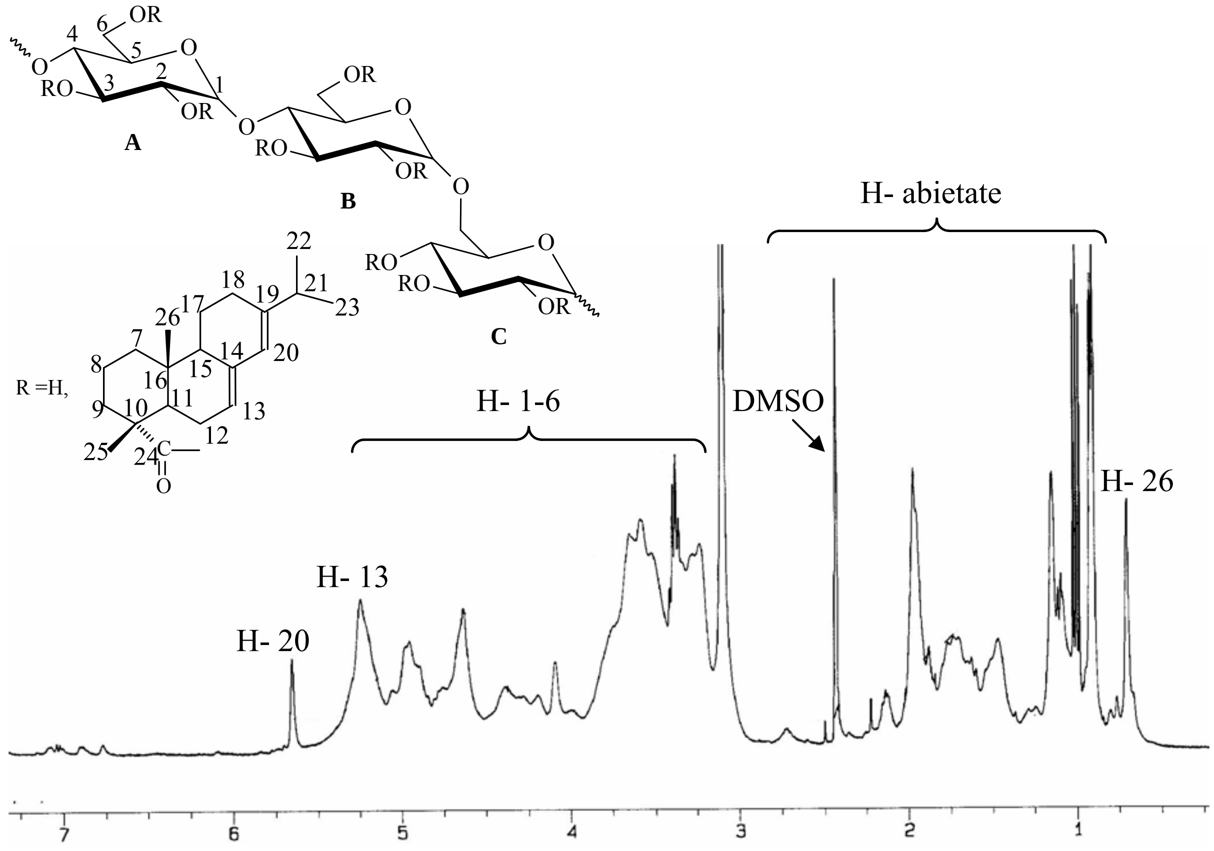


Fig. 2.34: ^{1}H NMR (DMSO-d_6, NS 16) spectrum of pullulan abietate **52** (DS 0.06)

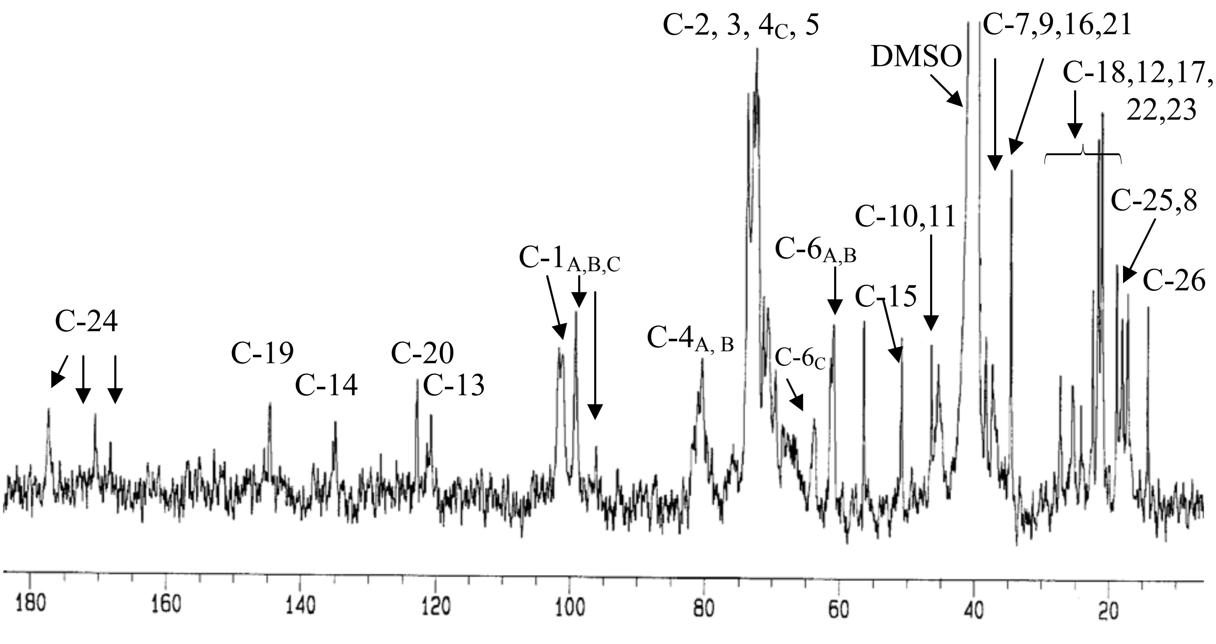


Fig. 2.35: ^{13}C NMR (DMSO-d_6, NS 7934) spectrum of pullulan abietate (sample **52**)

Esterification of pullulan with abietic acid was also carried out using *in situ* activation of abietic acid with *N,N`*-carbonyldiimidazole. The imidazolide of the abietic acid is formed as reactive species (see for details of this procedure in section 2.2.4). DS_{EA} 0.10 was achieved for sample **57** prepared by using molar ratio 1/1/1 (AGU/abietic acid/CDI). Sample **57** is water-soluble. This DS_{EA} value appeared comparable with the values obtained for pullulan abietates synthesised by *in situ* activation with Tos-Cl (sample **52**, DS_{Tit} 0.06). FTIR (KBr) spectrum of **57** is also similar to the FTIR spectrum of sample **52**.

A rather new method for the esterification of cellulose is the *in situ* activation of carboxylic acid with iminium chlorides, which has been reported recently (Hussian et al., 2004). Abietic acid was reacted with oxalyl chloride/DMF to yield iminium chloride of the abietic acid, which then reacts with pullulan. Pullulan abietate **58** was synthesised with DS_{EA} 0.07. FTIR (KBr) spectrum of **58** showed success of the reaction as ester carbonyl peak appeared at 1726 cm^{-1} and $C\text{-}O\text{-}C_{Ester}$ appeared at 1246 cm^{-1}. Both the values were comparable with FTIR spectra of other pullulan abietates (see for details of this procedure in section 2.1.4).

Gel permeation chromatography (GPC) was applied to obtain information about the degradation of the pullulan backbone during the conversion. DP values of pullulan abietates 32 (**56,** DS_{EA} 0.12), 95 (**57**, DS_{EA} 0.10) and 36 (**58**, DS_{EA} 0.07) were achieved if pullulan with Mr~100 000 is the starting polymer. Thus, esterification via imidazolide is much milder compared to conversion via *in situ* activation with tosyl chloride and iminium chloride.

Thermal decomposition temperatures (*T*d) 262°C was obtained from thermogravimetric analysis (TGA) for pullulan abietate **52** indicated the polymer obtained is thermally stable.

2.2.5.1. Self-assembly behaviour of pullulan and pullulan abietate and adsorption onto cellulose

It has become a dream of many scientists to develop a synthetic product that matches or surpasses the properties of wood. The structure of the cell wall in wood can be used as a guide for the development of a multiphase composite that exhibits a gradual transition between two distinct phases potentially leading to the development of a synthetic wood composite. Keeping this question in mind, through the study of the self-assembly behaviour of pullulan and pullulan derivatives, onto a model cellulose surface (Fig. 2.36) one can get further insight into the interactions between a hemicelluloses and cellulose present in the wood.

Self-assembly behaviour of pullulan abietate **51** is studied using surface plasmon resonance, which is capable of monitoring adsorption onto a biomimetic cellulose coated gold surface. It is important to note that cellulose itself has no self-assembly behaviour in aquous media. Therefore, timethylsilylated cellulose (TMSC) was used to prepare SAMs (self-assembled monolayers), which were transferred onto surface of gold by using langmuir-blodgett (LB) technique. In next step, desilylation was carried out to get regenerated cellulose surface (Schaub et al., 1993).

These studies of SAMs formation and adsorption onto cellulose surface were carried out in Virginia Polytechnic Institute and State University USA, in collaboration with professor Wolfgung G. Glasser.

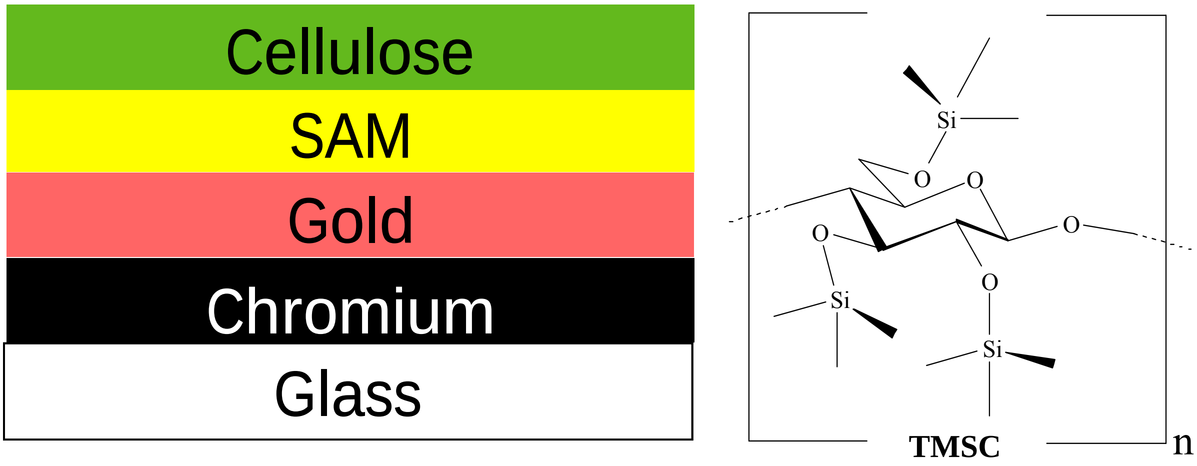


Fig. 2.36: Model cellulose surface (SAMs prepared after desilylation of TMSC)

There are several transfer modes for the deposition of molecules onto a substrate, X, Y, and Z-type transfer, which are shown in Fig. 2.37. As we used hydrophobic substrate, then most common transfer is Y-type has been followed (Petty et al., 1996). For Y-type deposition, the substrate is lowered into the sub-phase where the molecules orient their hydrophobic regions toward the substrate. On the upstroke, the polar head groups of molecules on the surface are attracted to the outward facing head groups already deposited on the substrate. The film is built up by continuous upward and downward strokes until an even number of layers is achieved with tails facing the air.

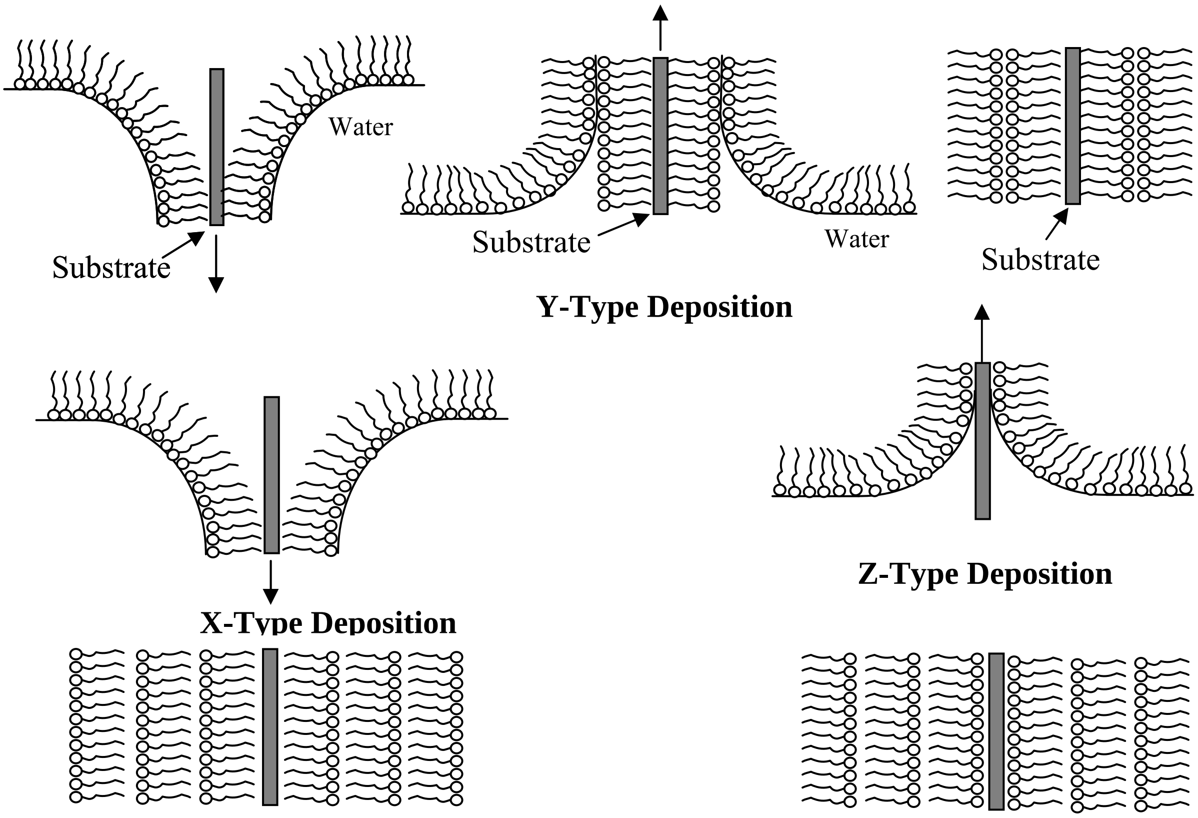


Fig. 2.37: Different LB-Deposition Modes

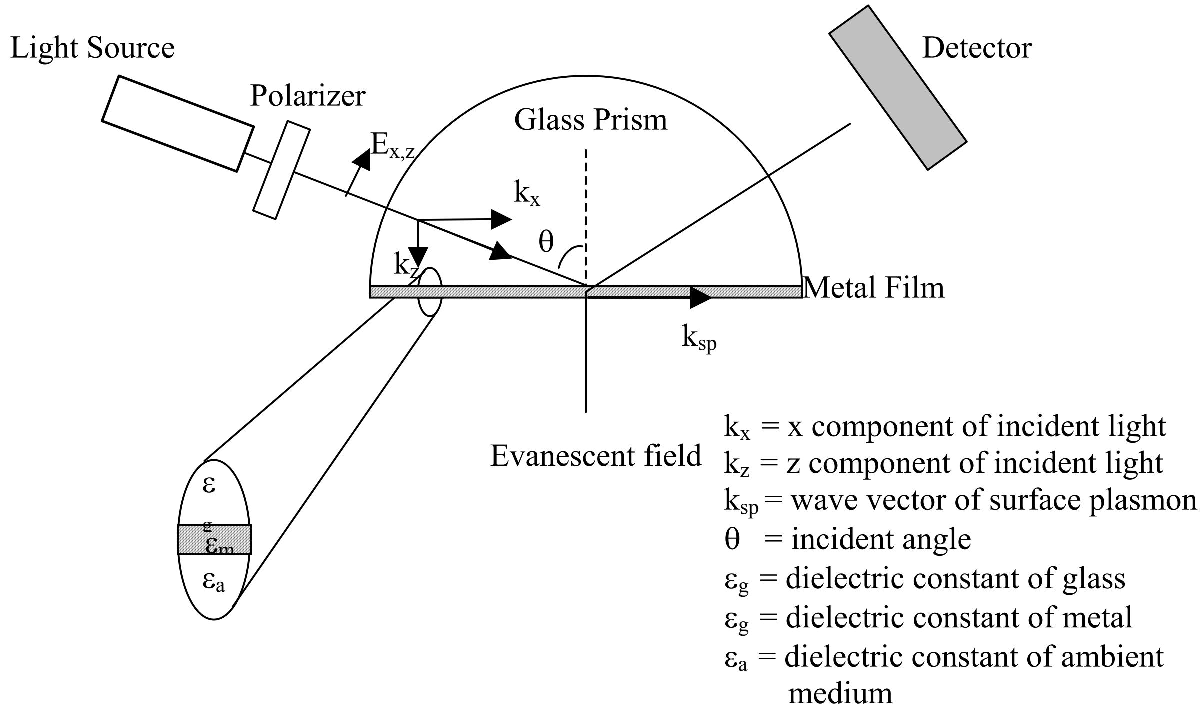


Fig. 2.38: Schematic of Kretschmann prism configuration

After transferring the cellulose surface over gold, pullulan and pullulan abietate **51** were transferred onto regenerated cellulose surface and adsorption was studied by surface plasmon resonance (SPR). SPR apparatus used is the Kretschman prism arrangement (see Fig. 2.38, Liedberg et al., 1998). Surface concentration (thickness of adsorbed layer = *t*) of adsorbed pullulan and sample **51** was determined by using following Feijter equation.

$$\Gamma = \frac{d\,(n_f - n_s)}{dn_s/dc} = \frac{\Delta\theta_a}{d\theta/dd}\,\frac{(n_f - n_s)}{dn_s/dc}$$

Γ : adsorbed molecules per unit area (mol/cm^2)
d : thickness of adsorbed film
n_f : refractive index of film
n_s : refractive index of bulk solution (without the adsorbent)
dn_s/dn_c : refractive index increment of adsorbent
$\Delta\theta$: change in angle corrected for bulk refractive index change
$d\theta/dd$: angular dependence on d of films with refractive index n_f

Samples of various known concentrations of pullulan and pullulan abietate **51** showed self-assembly behaviour from aqueous solution and adsorption onto cellulose surface. Change in refractive index θ_{SP} (Fig. 2.39) was observed for pullulan and pullulan abietate, which clearly indicates the adsorption of pullulan and of pullulan abietate onto the cellulose surface, which was the objective of this project.

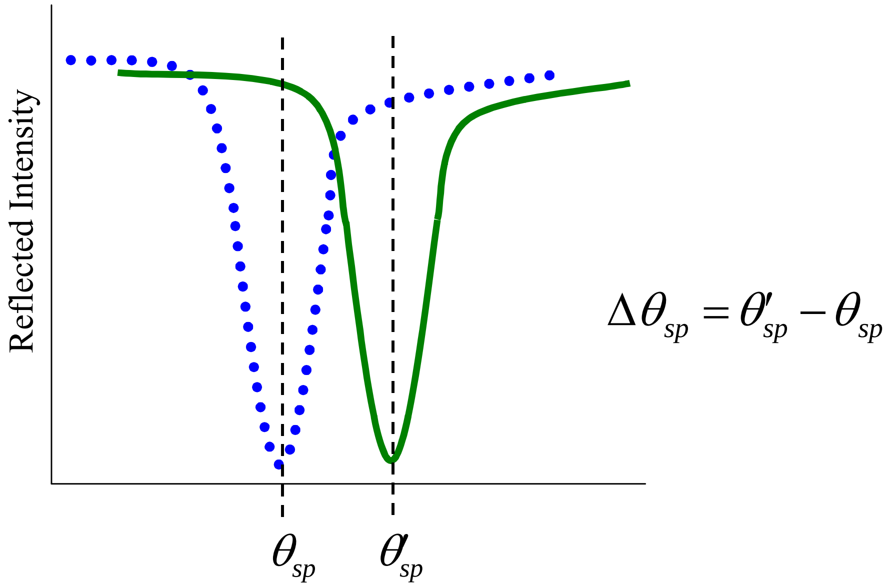
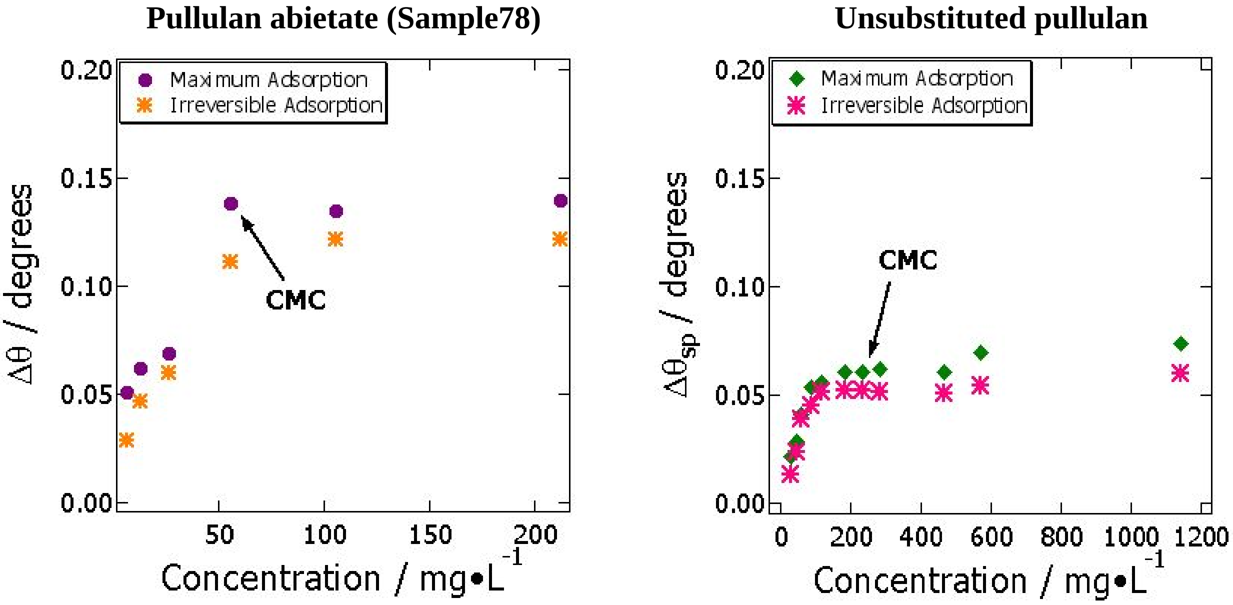


Fig. 2.39: Change in SPR output with adsorbed material

Hydrophobic abietic acid group arranged itself to minimize interactions with water. Micelles were formed as a result of this process, for this reason, hydrophilic core of pullulan surrounds the hydrophobic abietic acid groups. The concentration at which micelles initially formed is called critical micelle concentration (CMC). CMC was observed for unsubstituted pullulan at the concentration of 200 mg/L and CMC for pullulan abietate **51** was possible at 50 mg/L (Fig. 2.40).



Pullulan abietate (Sample78)

Unsubstituted pullulan

Fig. 2.40: Adsorption of unsubstituted pullulan and pullulan abietate **51** (DS 0.04) onto cellulose surface

Lower concentration for CMC was observed for pullulan abietate, which resulted from the hydrophobic behaviour of abietate moiety. On the other hand, both the pullulan and pullulan abietate show self-assembly onto a cellulose surface, however, self-assembly of pullulan occurs at a much lower concentration. Similar results are already reported for cholesteroyl pullulan (Akihiro et al., 1978; Akiyoshi et al., 1993).

AFM images were recorded for regenerated cellulose (cellulose II) surface, unsubstituted pullulan and pullulan abietate **51** (DS 0.02). AFM images revealed the adsorption of pullulan and pullulan abietate onto regenerated cellulose surface, which can be seen clearly in the AFM images (Fig. 2.41).

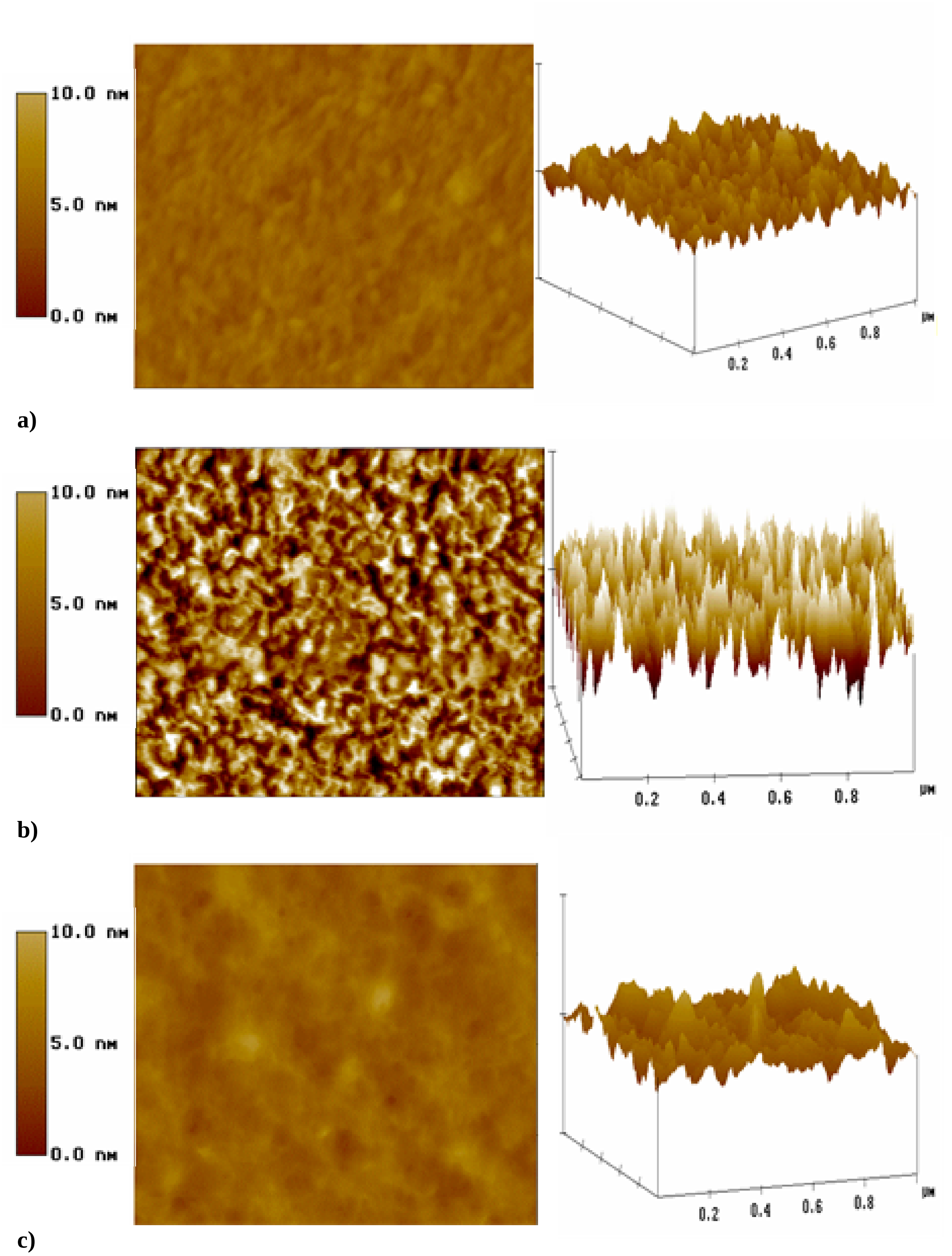


Fig. 2.41: AFM images of **a**) regenerated cellulose, roughness = 1.0 nm, **b**) unsubstituted pullulan, roughness = 1.4 nm and **c**) pullulan abietate **51** (DS 0.04), roughness = 2.6 nm

Adsorption onto cellulose is clearly indicated from the roughness value of the surface formed by the adsorption of pullulan or pullulan abietate. AFM results showed more pullulan abietate adsorbed onto cellulose surface rather than unsubstituted pullulan, as roughness value obtained for unsubstituted pullulan 1.4 nm is less then the roughness value obtained for pullulan abietate, i.e. 2.6 nm.

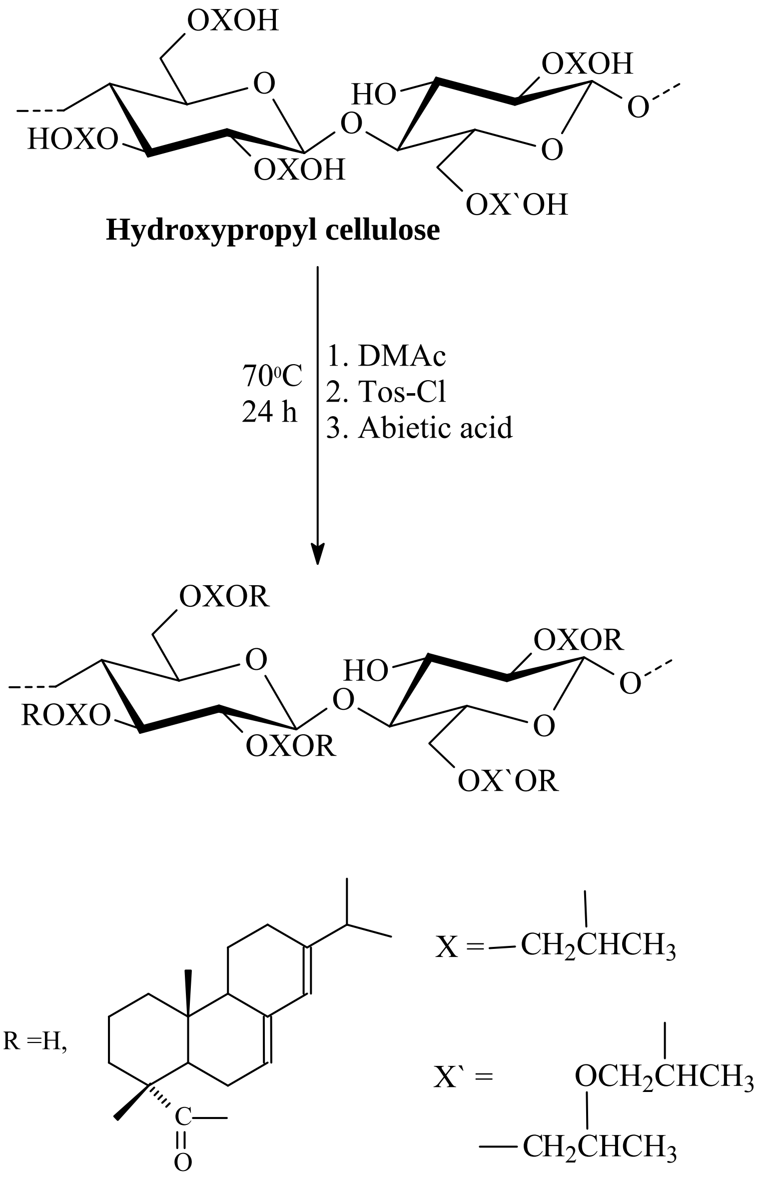
Summarising the results, one can conclude that both the pullulan and pullulan abietate show self-assembly onto a cellulose surface, however, self-assembly of pullulan occurs at a much lower concentration. Qualitatively, more pullulan abietate adsorbs onto cellulose surface in comparision with unsubstituted pullulan. Changes in refractive index above the CMC are due to a change in the bulk refractive index of the solution and must be accounted for in quantitative surface concentration calculations.

2.2.6. Synthesis of hydroxypropyl cellulose abietates with Tos-Cl in DMAc

Hydroxypropyl cellulose (HPC) is water-soluble and introduction of hydrophobic groups as ester can make the polymer amphiphilic in nature. Hydroxypropyl cellulose (HPC) is has broad spectrum of uses in food, pharmaceutical industry and film forming properties mainly due to it’s hydrophilic nature.

We are focusing on the synthesis and characterization of amphiphilic and novel HPC esters of abietic acid. Amphiphilic HPC abietate (**59-61**) were synthesised using *in situ* activation of abietic acid with Tos-Cl (Fig. 2.42). Results are summarized in Tab. 2.7. HPC dissolved in DMAc was allowed to react with abietic acid and Tos-Cl at 70°C and HPC abietates were synthesized using different molar ratios of AGU/abietic acid/Tos-Cl. Compound **59** and **60** were synthesized with lower DS 0.21 and DS 0.22 (Tab. 2.7) were soluble in usual organic solvents, however, sample **61** was prepared with DS 0.91 was only soluble in $CHCl_3$.

The mixed cellulose ether esters, i.e. HPC abietates were characterized using FTIR spectroscopy, EA and NMR spectroscopic studies. DS of the esters were calculated by using saponification and titration method. FTIR (KBr) spectrum of **61** showed two characteristic peaks typical for the ester moieties at about 1248 ν ($C\text{-}O\text{-}C_{Ester}$) cm^{-1} and 1735 ν (CO_{Ester}) cm^{-1} (Fig. 2.43). Similar FTIR spectra were obtained for all the products.



59-61

Fig. 2.42: Schematic plot of the conversion of hydroxypropyl cellulose with abietic acid applying *in situ* activation with Tos-Cl

Elemental analysis revealed the absence of sulphur in the samples showing that there is no remarkable introduction of tosylate groups neither covalently bounded nor as impurity. DS of samples **59-61** was calculated by EA and was found comparable to the DS calculated by titration method (see Tab. 2.7).

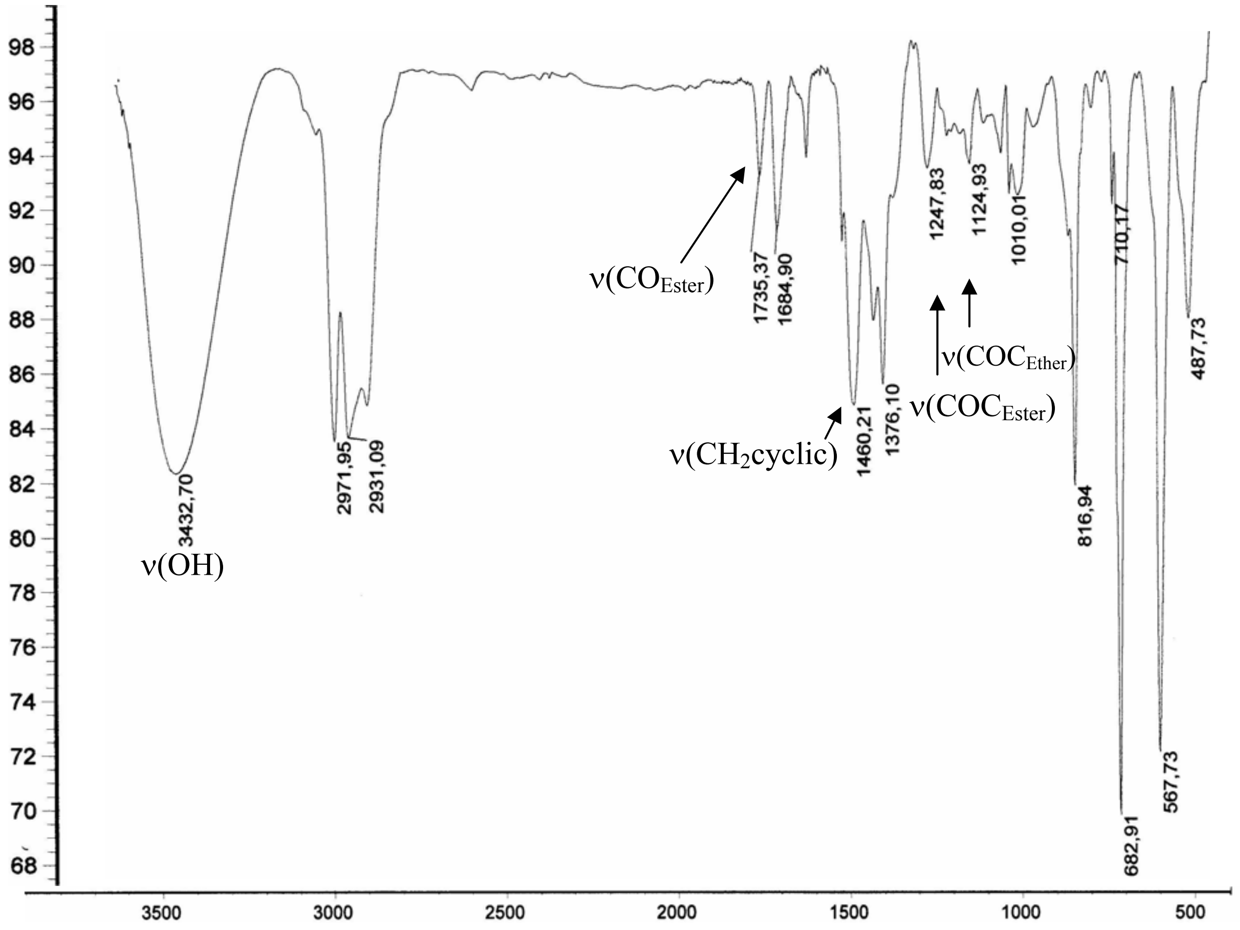


Fig. 2.43: FTIR (KBr, %transmittance) spectrum of hydroxypropyl cellulose abietate (sample **61**)

Tab. 2.7: Conditions and results of the reactions of HPC dissolved in DMAc with abietic acid after *in situ* activation with Tos-Cl

Samples	Molar ratio[a]	DS[b]	DS[c]	Yield (g)	Elemental Analysis		Solubility
					% C	% H	
59	1:0.25:0.25	0.21	0.14	5.26	52.50	9.66	DMSO, DMAc, THF, DMF, $CHCl_3$
60	1:0.5:0.5	0.22	0.16	5.20	53.23	9.88	DMSO, DMAc, THF, DMF, $CHCl_3$
61	1:1:1	0.91	0.99	8.35	51.00	9.06	$CHCl_3$

[a]) = HPC: Tos-Cl: abietic acid

[b]) = DS calculated by titration method

[c]) = DS calculated by EA

It was possible to completely functionalise remaining OH groups of hydroxypropyl cellulose abietate **61** by peracetylation using acetic anhydride in pyridine as solvent. FTIR (KBr) spectrum shows the absence of OH signals indicates the complete conversion of remaining OH groups by acetic anhydride. Increase in the intensity of CO_{Ester} signal at 1735 cm^{-1} was also observed. Furthermore, the peracetylated sample **61.1** (from starting polymer **61**) was not soluble in usual organic solvent.

^{13}C NMR spectrum ($CDCl_3$) of sample **61** recorded in $CDCl_3$ (Fig. 2.44) showed the characteristic carbonyl peak at δ = 174.0 ppm, which is indicative of introduction of ester function in the HPC backbone. No signal was found for abietic acid carbonyl. However, signals of cyclic rings are not possible to assign in spectrum due to less substitution and free rotation of abietic acid moiety as it is away from the HPC backbone.

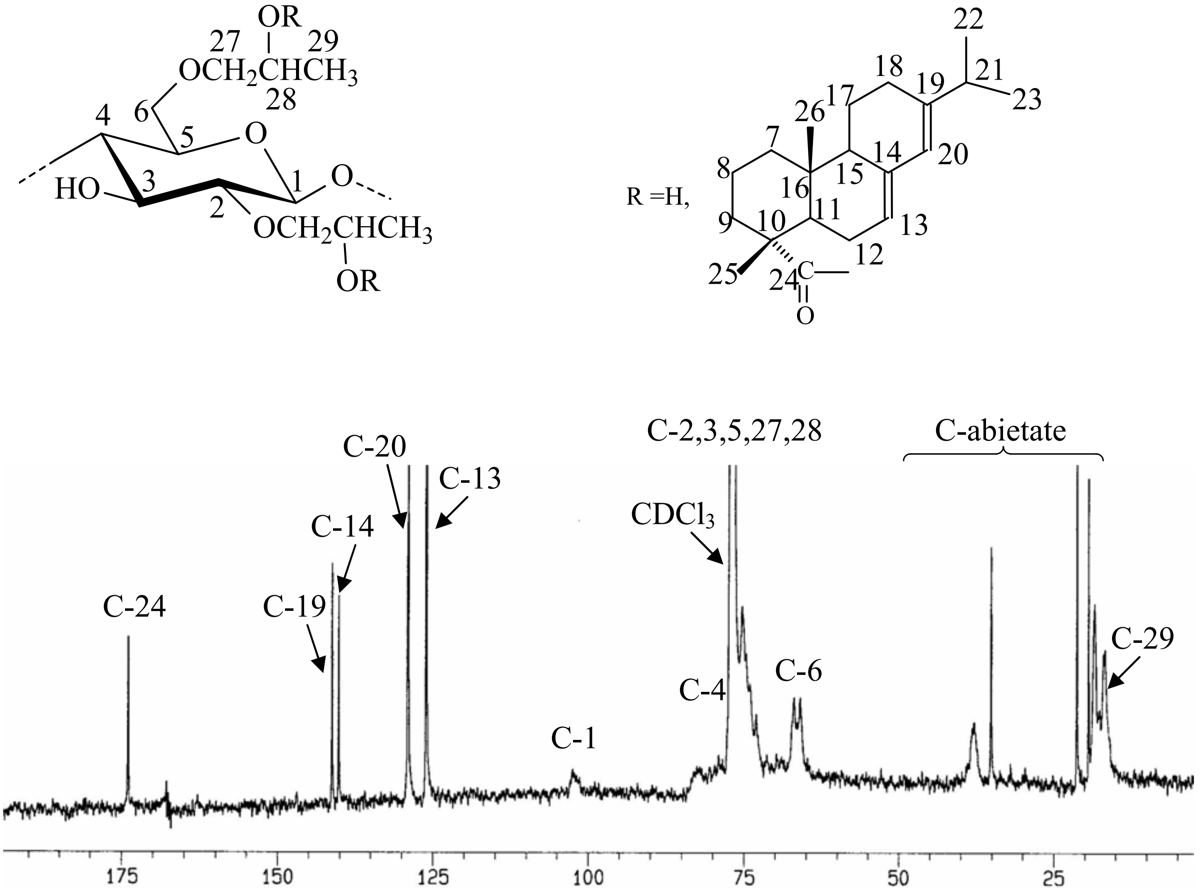


Fig. 2.44: ^{13}C NMR ($CDCl_3$, NS 20480) spectrum of hydroxypropyl cellulose abietate (sample **61**)

Well resolved AGU obtained showed signals at δ = 66.0-102.0, which are comparable with the parent polymer HPC. Signals of C-27 and C-28 are overlapped with the signals of AGU, however, methyl of hydoxypropyl absorbs at 17.2 ppm. Unsaturated carbons showed signals at δ = 126.0 (C-13), 129.0 (C-20), 140.2 (C-14) and 141.2 (C-19) ppm.

2.2.7. Synthesis of dextran abietates with Tos-Cl in DMAc/LiCl

Dextran dissolved in DMAc/LiCl was allowed to react with abietic acid using Tos-Cl as *in situ* activating agent. Dextran abietates **62-64** were synthesized with low DS by using different molar ratios of reactants using pyridine as base. Compound **62** and **63** were synthesized from dextran MW 30,000 and sample **64** was synthesized from dextran MW 70,000. Results are summarized in Tab. 2.8. Dextran abietates with low DS were characterized by FTIR spectroscopy, EA and NMR spectroscopic studies. FTIR (KBr) spectrum of **62** showed two characteristic peaks typical for the ester moieties at about 1238 cm^{-1} ($C\text{-}O\text{-}C_{Ester}$) and about 1715 cm^{-1} (CO_{Ester}). Similar FTIR spectra were obtained for all dextran abietates.

Elemental analysis revealed the absence of sulphur in the samples showing that there is no introduction of tosylate groups neither covalently bounded nor as impurity. DS of abietates was also possible to calculate with the help of elemental analysis, which appeared comparable to the values obtained by titration method. (see Tab. 2.8).

Tab. 2.8: Conditions and results of the reactions of dextran dissolved in DMAc/LiCl with abietic acid after *in situ* activation with Tos-Cl

Samples	Molar ratio[a]	DS[b]	DS[c]	Elemental analysis		Yield	Solubility
				% C	% H	(g)	
62	1:1:1:2	0.14	0.15	52.74	7.50	4.5	DMSO, DMAc, DMF
63	1:1.5:1.5:3	0.17	0.28	57.62	7.64	5.2	DMSO, DMAc, DMF, THF
64*	1:1.5:1.5:3		0.09	45.10	7.07	2.6	Water, DMF, DMAc, DMSO

* = Dextran MW 70,000 was used

[a]) = AGU:Tos-Cl: abietic acid: pyridine

[b]) = DS calculated by titration method

[c]) = DS calculated by EA

Regarding solubility of dextran abietates **62-64**, all samples were readily soluble in DMSO, DMAc and DMF. Sample **62** (DS 0.14) was additionally soluble in THF. Sample **64** (DS 0.09) synthesised from high molecular weight dextran was additionally water-soluble.

^{13}C NMR spectrum of **62** recorded in DMSO-d_6 shows the characteristic ester carbonyl peak at δ = 179.5 and 177.4 ppm, which indicates the introduction of abietic acid ester moiety. AGU absorbs at δ = 98.7-66.7 ppm. Unsaturated carbons showed signals at δ = 120.7 (C-13), 122.7 (C-20), 136.3 (C-14) and 144.7 (C-19) ppm (see Fig. 2.45).

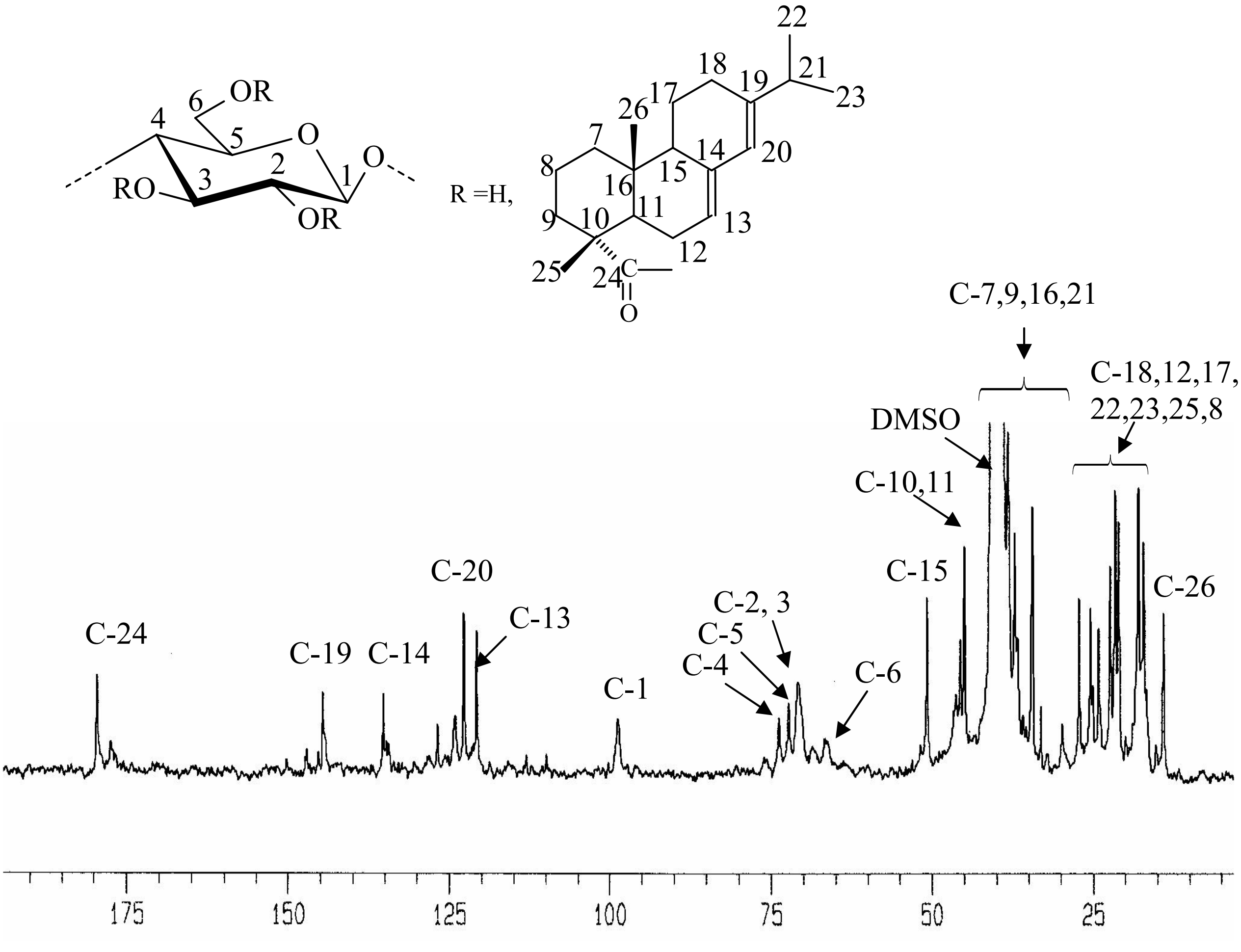


Fig. 2.45: ^{13}C NMR (DMSO-d_6, NS 50,000) spectrum of dextran abietate (sample **62**, DS 0.14)

Typical ^{1}H NMR (CDCl$_3$) spectrum of **62** (DS 0.14) after peracetylation showed well-resolved AGU at δ = 3.41-5.50 ppm (AGU-H). Protons of abietate moiety appeared in the range of 0.73-2.75 ppm. Unsaturated protons of abietate rings appeared at 5.71(H-20) and 5.31 (H-13, overlapped with AGU) ppm (Fig. 2.46).

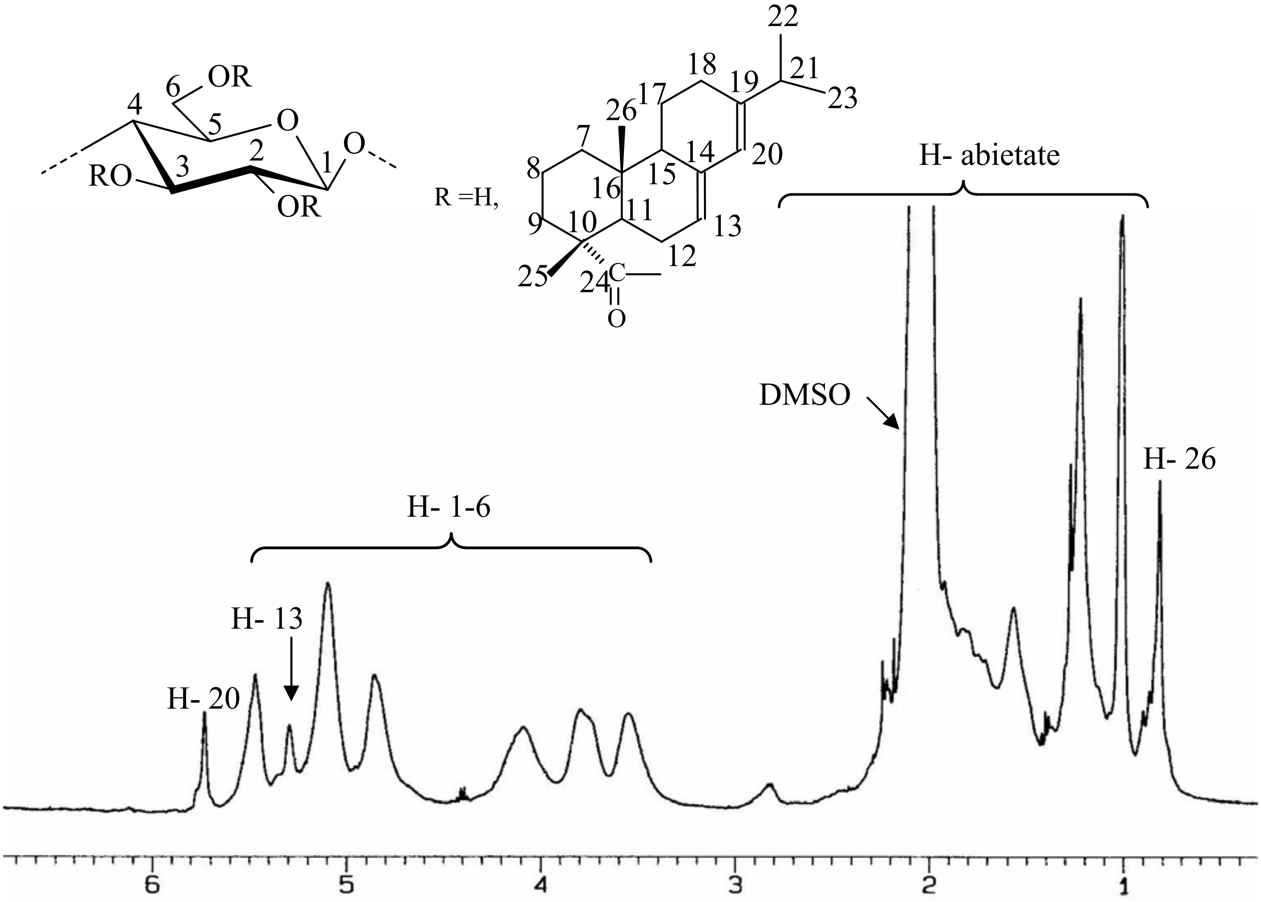


Fig. 2.46: ^{1}H NMR (CDCl$_3$, NS 16) spectrum of dextran abietate **62** (DS 0.14)

3. Experimental

3.1. Materials

Avicel (Fluka, “Avicel® PH-101”, DP 280) was used. Pullulan (70074), dextran MW 30,00 and dextran MW 70,000 polymers obtained from Fluka were dried under vacuum at 110°C for 8 h before use. LiCl was dried for 6 h at 105°C in vacuum prior to use. Pyridine was distilled over CaH_2. Cellulose acetate 2.5 (CA-398-3) Eastman® was obtained from Eastman Chemical Company. Hydroxypropyl cellulose was obtained from Hercules. All other chemicals supplied by Fluka, were used without further purification.

3.2. Measurements

^{13}C NMR spectra were acquired on a BRUKER AMX 400MHz spectrometer. The cellulose esters were measured in DMSO-d_6, $CDCl_3$ and THF-d_8 at 40°C and 70°C, respectively. The number of scans was in the range from 5,000 to 80,000.

^1H NMR spectra of the esters were acquired in $CDCl_3$ after peracylation of the unmodified hydroxyl groups (Heinze and Schaller, 2000) to determine the DS values. FTIR spectra were measured on a Bio-Rad FTS 25 PC using the KBr pellet technique.

Thermal decomposition temperatures of cellulose esters were determined by thermogravimetric analysis (TGA) on the Mettler Toledo TC 15 Mettler TG 50 Thermo balance. The thermal decomposition temperature (*T*d) was reported as the onset of significant weight loss from the heated sample. Samples (10 mg) were measured under air with a temperature increase of 10°C/min from 35°C up to 600°C.

Elemental analyses were performed by CHNS 932 Analyzer (Leco).

For GPC analysis, an equipment of JASCO was used including degasser (DG-980-50), pump (PU-980), RI-detector (RI-930) and UV-detector (UV-975) working at 254 nm. DMSO, Water & THF was used as eluent (30°C, 1 mL/min). The separation was carried out using columns from polymer standards service (Mainz, Germany) with 1,000, 10,000 and 1,000,000 Å. Polystyrene standards were used for calibration.

Preparation of gold slides for thin films of cellulose lipoates (Bartz et al., 2000): The glass slides (3.5 x 2.5 cm) were cleaned with aq.$NH_3/H_2O_2/H_2O$ (1:1:5) for 10 minutes at 80°C, washing with H_2O and isopropanol and dried in flow of N_2. These glass slides were coated

with gold using a Balzer BAE 250, vacuum coating unit under pressure of less than $5x10^{-6}$ hPa, typically depositing 50 nm of gold after first depositing 2 nm of Cr. The gold-coated glass slides were placed for 12 h in DMSO solution (2 mmol) of cellulose lipoate, rinsed with ethanol to remove unbound cellulose lipoate and dried in a stream of N_2.
LB-technique (LB-trough of Teflon^{TM}, Martin et al., 2002) used to transfer monomolecular film from surface of water to surface of substrate, i.e. TMSC coated on gold slides (Schaub et al., 1993). Surface pressure or adsorption of pullulan and pullulan abietate were calculated by Wilhelmy-Technique (Dynarowicz-Latka et al., 2001).
SPR measurements were performed in the kretschmann prism configuration (Liedberg et al., 1998) against ethanol. Optical coupling was achieved with a LASFN 9 prism, n= 1.85 at λ = 632.8 nm and index matching fluid n = 1.70 between prism and the BK270 glass sildes. The plasmon was excited with plane-polarized radiations using a He/Ne laser (632.6 nm, 5 mW).

3.3. Methods

3.3.1. Structural analysis of cellulose esters

3.3.1.1. Acetylation of CA 2.5 with acetic anhydride

1.0 g cellulose acetate 2.5 was dissolved in 10 mL pyridine. For complete acetylation, 10 mL acetic anhydride was added. The reaction mixture was heated up to 60°C for 24 h along with stirring. Isolation of the polymer **CA-2** was carried out by precipitation into 200 mL ethanol, washing with ethanol and drying in vacuum at 60°C.
Yield: 1.20 g
DS_{Acetat} = 2.96 (determined by means of ^1H NMR spectroscopy after perpropionylation)
FTIR (KBr): no ν (OH), 2890 ν (C-H), 1238 ν ($C\text{-}O\text{-}C_{Ester}$), 1750 ν ($CO_{Ester}$) cm^{-1}
^1H NMR ($CDCl_3$): δ (ppm) = 5.09 (H-3), 4.81 (H-2), 4.42 (H-1,6), 4.06 (H-6`), 3.73 (H-4), 3.56 (H-5), 2.14 (CH_3-6), 2.02(CH_3-2), 1.96 (CH_3-3)

Perpropionylation of ***CA-2*** *at 60°C*

Perpropionylation of **CA-2** was carried out by reacting 0.5 g of **CA-2** with 8 mL propionic anhydride and 8 mL pyridine for 24 h at 60°C in N_2 atmosphere under stirring. The polymer

was precipitated in 250 mL ethanol and washed with ethanol (250 mL) four times and then dried at 60°C under vacuum.
Yield: 0.45 g
DS_{Acetat} = 2.96 (determined by means of ^{1}H NMR spectroscopy)
FTIR (KBr): no ν (OH), 2890 ν (C-H), 1238 ν ($C\text{-}O\text{-}C_{Ester}$), 1750 ν ($CO_{Ester}$) cm^{-1}
^{1}H NMR (of perpropionate in $CDCl_3$): δ (ppm) = 5.00 (H-3), 4.73 (H-2), 4.33 (H-1,6), 3.99 (H-6`), 3.64 (H-4), 3.48 (H-5), 2.06 (CH_3-6), 1.94 (CH_3-2), 1.88 (CH_3-3)

Perpropionylation of ***CA-2*** *at 120°C*

Perpropionylation of **CA-2** was carried out by reacting 0.3 g of **CA-2** with 6 mL propionic anhydride and 6 mL pyridine for 24 h at 120°C in N_2 atmosphere under stirring. The polymer was precipitated in methanol and washed with methanol (250 mL) four times and then dried at 60°C under vacuum.
Yield: 0.20 g
DS_{Acetat} = 2.96 (determined by means of ^{1}H NMR spectroscopy)
FTIR (KBr): no ν (OH), 2890 ν (C-H), 1238 ν ($C\text{-}O\text{-}C_{Ester}$), 1750 ν ($CO_{Ester}$) cm^{-1}
^{1}H NMR ($CDCl_3$): δ (ppm) = 5.00 (H-3), 4.73 (H-2), 4.33 (H-1,6), 3.99 (H-6`), 3.64 (H-4), 3.48 (H-5), 2.06 (CH_3-6), 1.94 (CH_3-2), 1.88 (CH_3-3)

3.3.1.2. Propionylation of CA 2.5 with propionic anhydride (sample ***CA-1****)*

To calculate the DS of CA 2.5, perpropionylation was carried out. 1.0 g CA 2.5 dissolved in 10 mL pyridine was reacted with 10 mL propionic anhydride. The reaction mixture was heated up to 60°C for 24 h along with stirring under nitrogen. Isolation was carried out by precipitation into 200 mL ethanol, washing with ethanol and drying in vacuum at 60°C. This set of experiment was carried out twice and ^{1}H NMR spectra were recorded four times from each sample.
DS_{Acetat} = 2.32, 2,32, 2.35, 2.35 ($S^2 = 1.32 \times 10^{-4}$) and 2.37, 2,38, 2.37, 2.37 ($S^2 = 1.32 \times 10^{-4}$)

3.3.1.3. Trifluoroacetylation of CA 2.5 with trifluoroacetic acid/CDI

To prepare imidazolide of the trifluoroacetic acid, 3.0 g CDI was added in 20 mL DMSO followed by 1.43 mL trifluoroacetic acid. The mixture was stirred overnight at room temperature then added to the solution of 1.0 g CA 2.5 in 10 mL DMSO. The reaction mixture was stirred for 24 h at 80°C under N_2. The homogeneous reaction mixture was precipitated in 500 mL MeOH and the polymer was collected by filtration. After washing with 250 mL methanol three times, the polymer was dried at 60°C under vacuum to yield product **CA-3**.
Yield: 0.73 g
^{1}H NMR ($CDCl_3$): δ (ppm) = 3.42-5.09 (H-1-6), 2.06 (CH_3-6), 1.99 (CH_3-2), 1.93 (CH_3-3)

3.3.1.4. Nitrobenzoylation of CA 2.5 with 4-nitrobenzoic acid/CDI

3.09 g 4-nitrobenzoic acid dissolved in 20 mL DMSO followed by 3.0 g CDI to make its imidazolide and mixture was stirred over night. 1.0 g of re-precipitated CA 2.5 (from THF into EtOH) was dissolved in 20 mL DMSO. Both the mixtures were mixed and heated up to 60°C for 16 h along with stirring. Isolation of the polymer was carried out by precipitation into 200 mL ethanol. Polymer was washed with 200 mL ethanol three times and dried in vacuum at 60°C yielded product **CA-4**.
Yield: 1.05 g
DS = 2.60 (determined by means of ^{1}H NMR)
FTIR (KBr): no ν (OH), 2893 ν (C-H), 2959, 3100 ν (aromatic C-H), 1235 ν ($C\text{-}O\text{-}C_{Ester}$), 1531ν ($Ar\text{-}NO_2$), 1752 ν ($CO_{Ester}$) cm^{-1}
^{1}H NMR ($CDCl_3$): δ (ppm) = 5.00 (H-3), 4.73 (H-2), 4.33 (H-1,6), 3.98 (H-6`), 3.67 (H-4), 3.48 (H-5), 2.05 (CH_3-6), 1.94(CH_3-2), 1.87 (CH_3-3)

3.3.1.5. Nitrobenzoylation of CA 2.5 with 4-nitrobenzyl chloride

0.5 g CA 2.5 dissolved in 8 mL DMF was reacted with 1.5 g 4-nitrobenzyl chloride along with 12 mg DMAP. The reaction mixture was heated up to 60°C for 24 h along with stirring. Product **CA-5** was obtained by precipitation of the reaction mixture into 200 mL ethanol, washing with 200 mL ethanol thrice and drying in vacuum at 60°C.
Yield: 0.46 g
DS_{Acetat} = 2.66 (determined by means of ^{1}H NMR)

FTIR (KBr): no ν (OH), 2893 ν (C-H), 2957, 3115 ν (aromatic C-H), 1235 ν (C-O-C_{Ester}), 1532ν (Ar-NO_2), 1752 ν (CO_{Ester}) cm^{-1}
^{1}H NMR ($CDCl_3$): δ (ppm) = 5.09 (H-3), 4.82 (H-2), 4.45 (H-1,6), 4.06 (H-6`), 3.73 (H-4), 3.56 (H-5), 2.14 (CH_3-6), 2.02 (CH_3-2), 1.88 (CH_3-3), 8.42, 8.27, 8.01, 7.92 (H-aromatic)

3.3.1.6. Conversion of CA 2.5 with ethylisocyanato acetate

0.3 g CA 2.5 was allowed to react with 1 mL ethylisocyanato acetate in pyridine at 100°C under stirring. After 16 h 0.5 mL ethylisocyanato acetate was added and then stirred for another 24 h at 100°C. Polymer was isolated by precipitation in 250 mL diethyl ether and washing with 200 mL diethyl ether thrice followed by vacuum drying at 60°C yielded sample **CA-6**.
DS_{Acetat} = 2.43 (determined by means of ^{1}H NMR spectroscopy)
^{1}H NMR ($CDCl_3$): δ (ppm) = 5.09 (H-3), 4.82 (H-2), 4.42 (H-1,6), 3.98 (H-6`), 3.73 (H-4), 3.56 (H-5), 2.15 (CH_3-6), 2.02 (CH_3-2), 1.30 (CH_3-carbamate)

3.3.1.7. Conversion of CA 2.5 with phenylisocyanate

Phenylisocyanate 2.2 mL was added drop wise carefully in 1.0 g CA 2.5 dissolved in 10 mL pyridine. After removal of gases, the mixture was stirred for 12 h at room temperature. Polymer was precipitated in 250 mL MeOH. After washing with 200 mL methanol three times and dried under vacuum at 60°C, product **CA-7** was obtained.
DS_{Acetat} = 2.26 (determined by means of ^{1}H NMR spectroscopy)
FTIR (KBr): no ν (OH), 2956 ν (C-H), 3363 ν (aromatic C-H), 1232 ν (C-O-C_{Ester}), 1539ν (Ph-NO_2), 1753 ν (CO_{Ester}) cm^{-1}
^{1}H NMR ($CDCl_3$): δ (ppm) = 5.00 (H-3), 4.72 (H-2), 4.34 (H-1,6), 3.98 (H-6`), 3.71 (H-4), 3.49 (H-5), 2.06 (CH_3-6), 1.94 (CH_3-2), 1.87 (CH_3-3), 7.25, 7.20, 7.00 (H-aromatic)

3.3.2. Homogeneous cellulose esterification

3.3.2.1. Dissolution of cellulose in DMAc/LiCl

For a typical preparation, 1.0 g (6.2 mmol) of dried cellulose and 40 mL DMAc were kept at 120°C for 2 h under stirring. After the slurry has been allowed to cool down to 80°C, 3.0 g of anhydrous LiCl was added. The cellulose dissolved completely within 4 h by cooling down to room temperature under stirring.

3.3.2.2. Dissolution of cellulose in DMSO/TBAF

Avicel cellulose was simply dissolved by suspending 1.0 g of dried polymer in 66 mL DMSO and adding 6.6 g TBAF (Heinze *et al.*, 2000). Within 15 minutes a clear solution of cellulose was obtained.

3.3.2.3. Esterification of cellulose with lauric acid/Tos-Cl in DMAc/LiCl

To the solution of 4.0 g (25 mmol) of cellulose in DMAc/LiCl, 9.41 g (50 mmol) Tos-Cl were added, followed by 9.89 g (50 mmol) of lauric acid under stirring. The reaction mixture was stirred for 24 h at 80°C under N_2. The homogeneous reaction mixture was precipitated in 800 mL buffer solution (7.14 g K_2HPO_4 and 3.54 g KH_2PO_4 per litter of H_2O) and the polymer was collected by filtration. After washing the polymer with 800 mL water three times, Soxhlet extraction with ethanol was carried out for 24 h. The polymer was dried at 50°C under vacuum to yield product **4**.

Yield: 8.4 g (73%), white powder

DS = 1.55 (determined by means of ^{1}H NMR spectroscopy after peracetylation).

EA: 65% C, 8.14% H (results of EA and comparison of DS_{EA} and DS calculated by ^{1}H NMR are summarised in Tab.3.1)

FTIR (KBr): 3486 ν (OH), 2925, 2855 ν (CH), 1238 ν (COC_{Ester}), 1753 ν (CO_{Ester}) cm^{-1}

^{13}C NMR ($CDCl_3$): δ = 173.8 (CO), 104.0 (C-1), 102.6 (C-1`), 72.3 (C-2), 73.3 (C-3), 82.0 (C-4), 75.1 (C-5), 62.2 (C-6), 20.6-34.0 ($C_{Methylene}$), 13.9 (C_{Methyl}) ppm

Tab. 3.1: EA of cellulose esters mediated with Tos-Cl and comparison of DS values calculated by ^{1}H NMR spectroscopy and DS_{EA}

Sample No.	Carboxylic acids	Molar ratio[a]	Elemental analysis %		DS[b]	DS[c]
			C	H		
1	Capric	1:2:2:0	54.08	7.57	1.31	0.82
2	Caprylic	1:2:2:0	57.38	7.99	1.40	0.90
3	Decanoic	1:2:2:0	61.27	8.52	1.48	1.10
4	Lauric	1:2:2:0	65.51	9.70	1.55	1.38
5	Palmitic	1:2:2:0	69.04	10.79	1.60	1.45
6	Stearic	1:2:2:0	70.22	10.77	1.76	1.43
7	Caprylic	1:2:2:4	61.92	8.65	1.76	1.58
8	Lauric	1:2:2:4	64.94	9.62	1.79	1.30
9	Palmitic	1:2:2:4	70.19	10.50	1.71	1.68
10	Stearic	1:2:2:4	70.82	10.79	1.92	1.56
17	Caprylic	1:2:2:0	58.70	8.24	1.27	1.01
18	Lauric	1:2:2:0	64.80	9.59	1.55	1.27

[a]) = AGU:carboxylic acid:Tos-Cl:pyridine

[b]) = DS calculated by ^{1}H NMR spectroscopy after peracetylation

[c]) = DS calculated by EA

Peracetylation of sample ***4***

To determine the DS of cellulose laurate **4** by means of ^{1}H NMR spectroscopy, peracetylation of all unmodified hydroxyl groups was carried out. 2.0 g of **4** was allowed to react with 40 mL acetic anhydride and 40 mL pyridine in the presence of 50 mg of DMAP as catalyst for 24 h at 60°C in N_2 atmosphere under stirring. The polymer was precipitated in 500 mL distilled water, washed with ethanol (200 mL) four times and then dried at 50°C under vacuum to yield completely functionalised product **4.1** (see for result's details in Tab. 3.2)

Yield: 0.91 g

FTIR (KBr): no ν (OH), 1753 ν (CO_{Ester}) cm^{-1}

^{1}H NMR ($CDCl_3$): δ = 5.0 (H-3), 4.75 (H-2), 4.5 (H-1), 4.33 and 4.0 (H-6), 3.64 (H-4), 3.45 (H-5), 1.9 (H-20), 1.2-1.6 (H-10-17), 2.3 (H-8), 0.8 (H-18) ppm

Tab. 3.2: Synthesis of peracetylated cellulose esters of sample **1-22**: Conditions and solubility of the products

Acetylation mixture				Solubilities[d]				
Samples[a]	Amounts[b](g)	Reactants[c] (mL)	Yield (g)	DMSO	THF	DMF	Toluene	$CHCl_3$
1.1	0.5	10.0	0.45	+	-	+	-	+
2.1	0.5	10.0	0.48	-	-	-	-	+
3.1	2.0	40.0	2.11	-	-	-	-	+
4.1	2.0	40.0	1.40	-	-	-	-	+
5.1	2.0	40.0	1.58	-	-	-	-	+
6.1	2.0	40.0	1.91	-	+	-	+	+
7.1	2.0	40.0	2.06	-	-	-	-	+
8.1	2.0	40.0	1.56	-	+	-	-	+
9.1	2.0	40.0	2.06	-	+	-	+	+
10.1	2.0	40.0	1.93	-	+	-	-	+
11.1	0.5	10.0	0.24	+	-	+	+	+
12.1	0.5	10.0	0.55	+	-	+	-	+
13.1	1.0	20.0	0.92	+	-	+	-	+
14.1	2.0	40.0	1.67	-	-	-	-	+
15.1	2.0	40.0	1.12	-	-	-	-	+
16.1	2.0	40.0	1.57	-	+	-	+	+
17.1	1.0	20.0	0.50	+	-	+	-	+
18.1	2.0	40.0	1.44	-	-	+	-	+
19.1	2.0	40.0	1.69	-	-	+	-	-
20.1	1.0	20.0	1.06	+	-	+	-	+
21.1	2.0	40.0	1.92	-	-	-	-	+
22.1	2.0	40.0	2.01	-	-	-	-	+

[a]) = Resulting from cellulose esters **1-22**

[b]) = Amount of cellulose esters **1-22** taken for peracetylation

[c]) = Pyridine and acetic anhydride were taken in equal amounts along with a tip spatula of DMAP as catalyst

+) = Soluble

-) = Insoluble

Some products were precipitated in ethanol (**9.1, 11.1, 12.1 & 16.1**), methanol (**5.1, 6.1 & 20.1**) and water (**1.1-4.1, 7.1, 8.1, 10.1, 13.1-15.1,17.1-19.1, 21.1 & 22.1**).

The reaction conditions and solubilities of peracetylated cellulose esters **1.1-22.1** are summarized in Tab. 3.2.

DS of the ester moieties was calculated from ^{1}H NMR according to Goodlett et al., 1971 by equation;

$$DS = 3 - \frac{7 * I_{Acetyl}}{3 * I_{AGU}}$$

I_{acetyl} = peak integral of methyl protons of acetyl moieties.

I_{AGU} = peak integral of protons of anhydroglucose unit

Analytical data for the other cellulose esters prepared by in situ *activation of Tos-Cl in DMAc/LiCl*

Cellulose caprate (sample **1**)

FTIR (KBr): 3483 ν (OH), 2950 ν (CH), 1234 ν (COC_{Ester}), 1754 ν (CO_{Ester}) cm^{-1}

^{13}C NMR (THF-d_8): δ = 173.2 (CO), 104.2 (C-1), 102.2 (C-1\`), 73.6-77.5 (C-2, 3, 5), 82.8 (C-4), 63.6 (C-6), 20.6-34.5 ($C_{Methylene}$), 14.1 (C_{Methyl}) ppm

^{1}H NMR ($CDCl_3$) after peracetylation: δ = 5.05 (H-3), 4.75 (H-2), 4.5 (H-1), 4.2 (H-6a), 3.96 (H-6b), 3.62 (H-4), 3.49 (H-5), 1.9 (acetate methyl), 1.2-2.3 (caprate CH_2) and 0.8 (caprate methyl) ppm

Cellulose caprylate (sample **2**)

FTIR (KBr): 3473 ν (OH), 2930, 2958 ν (CH), 1234 ν (COC_{Ester}), 1754 ν (CO_{Ester}) cm^{-1}

^{13}C NMR ($CDCl_3$): δ = 173.5 (CO), 104.0 (C-1), 101.6 (C-1\`), 73.2-77.3 (C-2, 3, 5), 81.8 (C-4), 62.2 (C-6), 22.5-34.0 ($C_{Methylene}$), 14.0 (C_{Methyl}) ppm

^{1}H NMR ($CDCl_3$) after peracetylation: δ = 5.05 (H-3), 4.75 (H-2), 4.54 (H-1), 4.32 (H-6a) and 4.02 (H-6b), 3.63 (H-4), 3.50 (H-5), 1.9 (acetate methyl), 1.2-2.3 (caprylate CH_2) and 0.84 (caprylate methyl) ppm

Cellulose decanoate (sample **3**)

FTIR (KBr): 3480 ν (OH), 2926, 2856 ν (CH), 1240 ν (COC_{Ester}), 1754 ν (CO_{Ester}) cm^{-1}
^{13}C NMR ($CDCl_3$): δ = 173.5 (CO), 104.0 (C-1), 101.6 (C-1`), 73.3-77.2 (C-2, 3, 5), 81.9 (C-4), 62.4 (C-6), 22.6-34.0 ($C_{Methylene}$), 14.0 (C_{Methyl}) ppm
^{1}H NMR ($CDCl_3$) after peracetylation: δ = 5.07 (H-3), 4.79 (H-2), 4.54 (H-1), 4.32 (H-6a) and 4.05 (H-6b), 3.68 (H-4), 3.51 (H-5), 1.9 (acetate methyl), 1.2-2.3 (decanoate CH_2), 0.88 (decanoate methyl) ppm

Cellulose Stearate (sample **6**)

FTIR (KBr): 3482 ν (OH), 2924, 2854 ν (CH), 1239 ν (COC_{Ester}), 1754 ν (CO_{Ester}) cm^{-1}
^{13}C NMR ($CDCl_3$): δ = 173.1 (CO), 104.0 (C-1), 101.6 (C-1`), 73.2-77.3 (C-2, 3, 5), 81.9 (C-4), 62.5 (C-6), 22.6-34.0 ($C_{Methylene}$), 14.0 (C_{Methyl}) ppm
^{1}H NMR ($CDCl_3$) after peracetylation: δ = 5.06 (H-3), 4.79 (H-2), 4.54 (H-1), 4.38 (H-6a) and 4.05 (H-6b), 3.68 (H-4), 3.51 (H-5), 1.9 (acetate methyl), 1.2-2.3 (stearate CH_2) and 0.89 (stearate methyl) ppm

Cellulose palmitate (sample **9**)

FTIR (KBr): 3480 ν (OH), 2924, 2853 ν (CH), 1242 ν (COC_{Ester}), 1758 ν (CO_{Ester}) cm^{-1}
^{13}C NMR ($CDCl_3$): δ = 173.5 (CO), 103.6 (C-1), 101.6 (C-1`), 73.3-77.3 (C-2, 3, 5), 81.6 (C-4), 62.4 (C-6), 22.6-34.0 ($C_{Methylene}$), 14.0 (C_{Methyl}) ppm
^{1}H NMR ($CDCl_3$) after peracetylation: δ = 5.07 (H-3), 4.80 (H-2), 4.54 (H-1), 4.40 (H-6a) and 4.05 (H-6b), 3.68 (H-4), 3.51 (H-5), 1.9 (acetate methyl), 1.2-2.4 (palmitate CH_2), 0.89 (palmitate methyl) ppm

3.3.2.4. Synthesis of cellulose α-lipoate with α-lipoic acid/Tos-Cl in DMAc/LiCl

To the solution of 1.0 g of cellulose in DMAc/LiCl, 3.53 g Tos-Cl was added, followed by 3.82 g of α-lipoic acid under stirring. The reaction mixture was stirred for 16 h at 60°C under N_2. The homogeneous reaction mixture was precipitated in 600 mL EtOH, washed with 250 mL EtOH three times and the polymer was collected by filtration. The polymer was dried at 50°C under vacuum to yield product **23**.

Yield: 2.50 g (93%), yellowish powder

DS_{EA} = 1.45

EA: 48.20% C, 6.04% H, 21.32% S

FTIR (KBr): 3481 ν (OH), 2931, 2862 ν (CH), 1238 ν (COC_{Ester}), 1742 ν (CO_{Ester}) cm^{-1}

3.3.2.5. Synthesis of cellulose α-lipoate with α-lipoic acid/CDI in DMAc/LiCl

1.5 g CDI was dissolved in 30 mL DMAc followed by 1.91 g α-lipoic acid to obtain imidazolide of the α-lipoic acid. The mixture was stirred overnight then added to the solution of 1.0 g cellulose dissolved in DMAc/LiCl. The reaction mixture was stirred for 16 h at 60°C under N_2. The homogeneous reaction mixture was precipitated in 500 mL acetone and the polymer was collected by filtration. After washing with 250 mL acetone three times, the polymer was dried at 50°C under vacuum to yield product **25**.

Yield: 1.31 g, 99%

DS_{EA}: 0.18

EA: 34.81% C, 6.27% H, 5.94% S (results of EA are summarized in Tab. 2.2)

FTIR (KBr): 3469 ν (OH), 2919 ν (C-H), 1234 ν ($C\text{-}O\text{-}C_{Ester}$), 1731 ν ($CO_{Ester}$) cm^{-1}

Perpropionylation of cellulose α-lipoate **25**

Perpropionylation of all unmodified hydroxyl groups was carried out. For this purpose, 0.6 g of sample **25** was allowed to react with 8.0 mL propionic anhydride and 8.0 mL pyridine in the presence of 20 mg of DMAP as catalyst for 24 h at 60°C in N_2 atmosphere under stirring. The polymer was precipitated and washed with 250 mL ethanol four times and then dried at 60°C under vacuum to yield product **25.1**.

Yield: 0.16 g

FTIR (KBr): no ν(OH), 2984, 2946, 2889 ν(CH), 1738 ν(CO_{Ester}), 1440 (Cyclic C-H bending vibrations) cm^{-1}

^1H NMR ($CDCl_3$) after perpropionylation: δ (ppm) = 5.01 (H-3), 4.85 (H-2), 4.31 and 3.96 (H-6), 3.62 (H-4), 3.46 (H-5), 2.10-2.16 (H-7, 14), 3.04-3.13 (H-12, 13), 1.35-1.88 (H-8, 10), 0.99, 1.08, 1.18 (H-15 at C-3, 2, 6 respectively)

3.3.2.6. Esterification of cellulose with 4-nitrobenzoic acid/OX-Cl/DMF in DMAc/LiCl

To make iminium chloride of the 4-nitrobenzoic acid, 30 mL DMF was cooled at –20°C using dry ice, and then 1.76 mL oxalyl chloride was added drop wise very carefully. After gas-formation had stopped the 3.09 g 4-nitrobenzoic acid was added and mixed for 15 minutes at same temperature. The mixture was added to the solution of 1.0 g cellulose in DMAc/LiCl. The reaction mixture was stirred for 16 h at 60°C under N_2. The homogeneous reaction mixture was precipitated in 500 mL EtOH and the polymer was collected by filtration. After washing of the polymer with 250 mL EtOH three times, the polymer was dried at 50°C under vacuum to yield product **39**.

Yield: 1.02 g, 42%, white powder
DS: 0.94 (determined by means of ^{1}H NMR spectroscopy after perpropionylation)
EA: 44.86% C, 4.99% H, traces of chlorides (results of EA are summarized in Tab. 3.3)
FTIR (KBr): 3457 ν (OH), 2895 ν (C-H), 1239 ν (C-O-C$_{\text{Ester}}$), 1729 ν (CO$_{\text{Ester}}$) cm^{-1}

Tab. 3.3: EA of cellulose esters prepared by iminium chlorides and comparison of DS values calculated by ^{1}H NMR spectroscopy and by EA

Sample No.	Carboxylic acids	Molar ratio[a]	Elemental analysis %			DS[b]	DS[c]
			C	H	Cl		
28	Stearic acid	1:1:1	39.38	6.69	Traces	0.15	0.16
29	Stearic acid	1:2:2	52.68	7.14	2.75	0.21	0.18
30	Stearic acid	1:3:3	52.29	8.97	2.89	0.63	0.44
33	Admantane-1 carboxylic acid	1:1:1	40.98	6.72	Traces	0.47	0.25
35	Admantane-1 carboxylic acid	1:3:3	57.92	7.17	2.27	1.20	0.60
36	Admantane-1 carboxylic acid	1:6:6	53.63	7.12	2.66	0.66	0.57

[a]) = AGU:carboxylic acid:oxalyl chloride

[b]) = DS calculated by ^{1}H NMR spectroscopy after peracylation

[c]) = DS calculated by EA

Perpropionylation of ***39***

A mixture of 6.0 mL pyridine, 6.0 mL propionic acid anhydride and 50 mg DMAP was added to 0.3 g of the cellulose 4-nitrobenzoate **39**. After 24 h stirring at 80°C the reaction mixture was precipitated in 250 mL ethanol. For purification the isolated polymer was re-precipitated from chloroform into 100 mL ethanol, filtered off, washed with ethanol and dried in vacuum at 50°C yielded completely functionalized polymer **39.1**.

Yield: 0.4 g

DS = 0.94 (determined by means of ^{1}H NMR spectroscopy)

FTIR (KBr): no ν(OH), 2985, 2946, 2886 ν(CH), 1756, ν(CO_{Ester}) cm^{-1}

^{1}H NMR ($CDCl_3$): δ = 5.04 (H-3), 4.73 (H-2), 4.5 (H-1), 4.35 and 4.0 (H-6), 3.63 (H-4), 3.46 (H-5), 7.78-8.30 (H-7-8), 2.1 (H-9), 0.81 (H-10) ppm

3.3.2.7. Synthesis of cellulose furoate with 2-furan carboxylic acid/CDI in DMSO/TBAF

3.0 g CDI was dissolved in 30 mL DMSO followed by 2.07 g 2-furan carboxylic acid to obtain imidazolide of the 2-furan carboxylic acid. The mixture was stirred overnight then added to the solution of 1.0 g cellulose dissolved in DMSO/TBAF. The reaction mixture was stirred for 24 h at 80°C under N_2. The homogeneous reaction mixture was precipitated in 500 mL EtOH and the polymer was collected by filtration. After washing with 250 mL EtOH three times, the polymer was dried at 50°C under vacuum to yield product **50**.

Yield: 1.40 g, 61%

DS 1.91: (determined by means of ^{1}H NMR spectroscopy after perpropionylation)

FTIR (KBr): 3493 ν (OH), 3142 (C-H aromatic), 2892 ν (C-H), 1233 ν (C-O-C_{Ester}), 1579 ν (aromatic furan ring), 1728 ν (CO_{Ester}) cm^{-1}

^{13}C NMR (DMSO-d_6): δ (ppm) = 157.3 (CO), 102.9 (C-1), 99.8 (C-1`), 72.3 (C-2), 73.9 (C-3), 80.2 (C-4), 76.3 (C-5), 63.1 (C-6), 143.4 (C-8), 118.8 (C-9), 112.1 (C-10), 147.6 (C-11)

Perpropionylation of cellulose furoate ***50***

To determine the DS of cellulose esters by means of ^{1}H NMR spectroscopy, perpropionylation of all unmodified hydroxyl groups was carried out. For this purpose, 0.6 g of sample **50** dissolved in 15 mL pyridine was allowed to react with 15 mL propionic anhydride in the presence of 20 mg of DMAP as catalyst for 24 h at 60°C in N_2 atmosphere under stirring. The polymer was precipitated and washed with 250 mL ethanol four times and then dried at 60°C under vacuum yielded product **50.1**.

Yield: 0.21 g

FTIR (KBr): no ν(OH), 2926, 2855 ν(CH), 1757 ν(CO_{Ester}) cm^{-1}

^1H NMR ($CDCl_3$): δ (ppm) = 5.00 (H-3), 4.85 (H-2), 4.38 and 4.08 (H-6), 3.66, 3.63 (H-4, 5), 6.50 (H-10), 7.20 (H-9), 7.56 (H-11), 2.04 (CH_2-propionate), 0.77, 0.93 (CH_3-2, 3-propionate)

Analytical data for cellulose esters synthesized by CDI method in DMSO/TBAF

Cellulose acetate (sample **66**)

FTIR (KBr): 3480 ν (OH), 2902 ν (C-H), 1238 ν (C-O-C_{Ester}), 1745 ν (CO_{Ester}) cm^{-1}

^{13}C NMR (DMSO-d_6): δ (ppm) = 170.1, 169.3 (CO), 102.65 (C-1), 100.1 (C-1`), 71.8-75.5 (C-2, 3, 5), 79.54 (C-4), 62.75 (C-6), 20.54 (CH_3)

^1H NMR (after perpropionylation in $CDCl_3$): 2.04 (CH_3), 3.08-5.47 (AGU)

Cellulose propionate (sample **67**)

FTIR (KBr): 3480 ν (OH), 2902 ν (C-H), 1238 ν (C-O-C_{Ester}), 1745 ν (CO_{Ester}) cm^{-1}

^{13}C NMR (DMSO-d_6): δ (ppm) = 170.1, 169.3 (CO), 102.65 (C-1), 100.1 (C-1`), 71.8-75.5 (C-2, 3, 5), 79.54 (C-4), 62.75 (C-6), 20.54 (CH_3)

^1H NMR (after perpropionylation in $CDCl_3$): 2.04 (CH_3), 3.08-5.47 (AGU)

Cellulose adamantate (sample **49**)

FTIR (KBr): 3458 ν(OH), 2910, 2855 ν(CH), 1728 ν(C=O_{Ester}) cm^{-1}

^{13}C NMR (DMSO-d_6): δ = 176.4 (CO), 102.6 (C-1), 99.5 (C-1'), 78.8 (C-4), 73.4 (C-3, C-5, C-2), 62.9 (C-6s), 61.6 (C-6), 40.1 (α-C), 38.8 (β-CH_2), 36.4 (δ-CH_2), 27.8 (γ-CH) ppm

^1H NMR ($CDCl_3$) after perpropionylation: δ (ppm) = 5.05 (H-3), 4.72 (H-2), 4.31 and 3.96 (H-6), 3.61 (H-4), 3.45 (H-5), 2.13 (CH_2-propionate), 2.31, 1.99, 1.91, 1.83, 1.67 (H-adamantane), 0.96, 1.11 (CH_3-2, 3-propionate)

3.3.3. Homogeneous synthesis of pullulan abietates

3.3.3.1. Dissolution of pullulan in DMAc

5.0 g pullulan was added in 100 mL DMAc. Mixture was stirred at 80°C for 30 minutes to get optical clear solution of pullulan.

3.3.3.2. Synthesis of pullulan abietate with abietic acid/Tos-Cl

To the solution of 5.0 g pullulan in DMAc, 2.5 mL pyridine base was added followed by 5.9 g Tos-Cl and 9.33 g abietic acid under stirring. The reaction mixture was stirred for 24 h at 70°C under N_2. The homogeneous reaction mixture was precipitated in 1.0 L distilled water and washed with 250 mL EtOH three times. The polymer was dried at 60°C under vacuum to yield product **52**.
Yield: 5.5 g
DS_{Tit} = 0.06
EA: 51.79% C, 7.26% H
FTIR (KBr): 3416 ν (OH), 2931 ν (C-H), 1724 ν (CO_{Ester}), 1246 ν ($C\text{-}O\text{-}C_{Ester}$) cm^{-1}
^{13}C NMR (DMSO-d_6): δ=177.4, 170.5, 168.1 (CO), 101.7, 99.3, 96.0 (C-1), 80.4 (C-$4_{A, B}$), 60.9-73.8 (C-2, 3, 4_C, 5, 6), 14.25 (C-26), 17.2-18.9 (C-25, 8), 21.1-27.5 (C-22, 23, 17, 12, 18), 34.6-38.3 (C-9, 7, 16, 21), 46.4 (C-10, 11), 50.9 (C-15), 120.8 (C-13), 122.9 (C-20), 134.9 (C-14) and 144.7 (C-19) ppm
^{1}H NMR (DMSO-d_6): δ = 3.26-5.27 (AGU-H), 0.73-2.75 (abietate moiety-H) and 5.71 (H-20) and 5.31 (H-13, overlapped with signals of AGU) ppm

3.3.3.3. Synthesis of pullulan abietate with abietic acid/CDI

1.86 g abietic acid dissolved in 30 mL DMF followed by 1.5 g CDI to make its imidazolide and mixture was stirred over night. The reaction mixture was added to 1.0 g pullulan dissolved in DMAc. The reaction mixture heated up to 70°C for 24 h along with stirring. The homogeneous reaction mixture was precipitated in 500 mL ethanol and washed with 200 mL ethanol thrice. The polymer was dried at 60°C under vacuum to yield product **57**.

Yield: 0.71 g
$DS_{EA} = 0.10$
EA: 43.38% C, 7.20% H
FTIR (KBr): 3404 ν (OH), 2926 ν (C-H), 1724 ν (CO_{Ester}), 1244 ν ($C\text{-}O\text{-}C_{Ester}$) cm^{-1}

3.3.3.4. Synthesis of pullulan abietate with abietic acid/Oxalyl chloride/DMF

To make iminium chloride of the abietic acid, 30 mL DMF was cooled at -20°C using dry ice, and then 0.59 mL oxalyl chloride was added drop wise very carefully. After gas-formation had stopped, 1.86 g abietic acid was added and mixed for 15 minutes at same temperature. The mixture was added to the solution of 1.0 g pullulan in DMAc. The reaction mixture was stirred for 24 h at 70°C under N_2. The homogeneous reaction mixture was precipitated in 500 mL acetone and the polymer was collected by filtration. After washing with 250 mL acetone three times, the polymer was dried at 50°C under vacuum to yield product **58**.
Yield: 0.70 g
$DS_{EA} = 0.18$
EA: 40.83% C, 7.14% H
FTIR (KBr): 3411 ν (OH), 2929 ν (C-H), 1726 ν (CO_{Ester}), 1246 ν ($C\text{-}O\text{-}C_{Ester}$) cm^{-1}

3.3.4. Homogeneous synthesis of hydroxypropyl cellulose abietates

3.3.4.1. Dissolution of hydroxypropyl cellulose in DMAc

5.0 g hydroxypropyl cellulose was added in 125 mL DMAc. Mixture was stirred at 110°C for 30 minutes to get completely dissolved and transparent solution of hydroxypropyl cellulose.

3.3.4.2. Synthesis of hydroxypropyl cellulose abietate with abietic acid/Tos-Cl

3.26 g Tos-Cl was added to the solution of 5.0 g hydroxypropyl cellulose in DMAc followed by 5.17 g abietic acid under stirring. The reaction mixture was stirred for 24 h at 70°C under N_2. The homogeneous reaction mixture was precipitated in 500 mL diethyl ether and washed with 250 mL Et_2O three times. The polymer was dried at 60°C under vacuum to yield the sample **61**.

Yield: 8.35 g
$DS_{Tit} = 0.91$
EA: 50.81% C, 8.43% H, $DS_{EA} = 0.99$
FTIR (KBr): 3433 ν (OH), 2931, 2972 ν (C-H), 1735 ν (CO_{Ester}), 1248 ν ($C\text{-}O\text{-}C_{Ester}$) cm^{-1}
^{13}C NMR ($CDCl_3$): δ = 174.0 (CO), 101.5 (C-1), 82.2 (C-4), 65.9, 66.9 (C-6), 72.4-79.1 (C-2, 3, 5, 27, 28), 16.8 (C-29), 17.2-37.7 (C-abietate), 141.2 (C-19), 140.2 (C-14), 129.0 (C-20), 126.1 (C-13) ppm

Peracetylation of sample ***61***

Peracetylation of all unmodified hydroxyl groups of cellulose hydroxypropyl abietate **61** was carried out. 2.0 g of **61** was allowed to react with 40 mL acetic anhydride and 40 mL pyridine in the presence of 50 mg of DMAP as catalyst for 24 h at 60°C in N_2 atmosphere under stirring. The polymer was precipitated in 500 mL ethanol, washed with ethanol (200 mL) four times and then dried at 50°C under vacuum to yield completely functionalised product **61.1**.
Yield: 1.20 g
FTIR (KBr): no ν (OH), 1739 ν (CO_{Ester}) cm^{-1}

3.3.5. Homogeneous synthesis of dextran abietates

3.3.5.1. *Dissolution of dextran in DMAc/LiCl*

5.0 g dextran (MW 30,000) was added in 100 mL DMAc and the mixture was stirred at 120°C for 2 h. After cooling the slurry at 80°C, 7.50 g LiCl was added. The polymer was completely dissolved within 10 minutes simply by stirring at room temperature. Similarly, dextran MW 70,000 can be dissolved in DMAc/LiCl as mentioned above, however, it takes 4 h after adding LiCl to become completely soluble at room temperature.

3.3.5.2. Synthesis of dextran abietate with abietic acid/Tos-Cl

To the solution of 5.0 g dextran dissolved in DMAc/LiCl, 3.7 mL pyridine was added followed by 8.82 g Tos-Cl and 14.0 g abietic acid under stirring. The reaction mixture was stirred for 24 h at 70°C under N_2. The homogeneous reaction mixture was precipitated in 800

mL diethyl ether and washed with 250 mL diethyl ether three times. The polymer was dried at 60°C under vacuum to yield product **62**.
Yield: 5.2 g
$DS_{Tit} = 0.17$
EA: 57.62% C, 7.64% H, $DS_{EA} = 0.28$
FTIR (KBr): 3398 ν (OH), 2928 ν (C-H), 1715 ν (CO_{Ester}) cm^{-1}, 1238 ν ($C\text{-}O\text{-}C_{Ester}$)
^{13}C NMR (DMSO-d_6): δ = 179.5, 177.4 (CO), 98.7 (C-1), 76.1 (C-4), 73.8 (C-5), 72.3 (C-2)-70.9 (C-3), 66.7 (C-6), 17.16 (C-26), 18.04-51.0 (C-abietate moiety), 120.7 (C-13), 122.7 (C-20), 135.3 (C-14) and 144.7 (C-19) ppm

Peracetylation of sample ***62***

Peracetylation of all unmodified hydroxyl groups of dextran abietate **62** was carried out. 2.0 g of **62** was allowed to react with 40 mL acetic anhydride and 40 mL pyridine in the presence of 50 mg of DMAP as catalyst for 24 h at 50°C in N_2 atmosphere under stirring. The polymer was precipitated in 400 mL ethanol, washed with ethanol (250 mL) four times and then dried at 50°C under vacuum to yield completely functionalised product **62.1**.
Yield: 1.20 g
FTIR (KBr): no ν (OH), 1754 ν (CO_{Ester}) cm^{-1}
^1H NMR ($CDCl_3$) after peracetylation: δ = 3.41-5.05 (AGU-H), 0.73-2.75 (abietate moiety-H) and 5.72 (H-20) and 5.45 (H-13) ppm

4. Summary

Structure elucidation of cellulose acetates (CA), different synthesis paths, analysis strategies and correlation of these structural features were studied. Alternative paths for the synthesis of CA were studied focusing on *in situ* activation of acetic acid. A number of different reaction paths were used to completely functionalize cellulose acetate 2.5 using different reactive intermediates, i.e. acetylation, propionylation, trifluoroacetylation, nitrobenzylation, ethyl carbanilation and phenyl carbanilation. Strategies for structure analysis by mean of ^{1}H NMR spectroscopy were discussed. The structures obtained were analyzed both on the level of the anhydroglucose unit (AGU) and along the polymer chain. No hints for a non-statistic distribution of the acetyl-groups along the polymer were observed. The esters synthesized were characterized in detail with regard to the DS, DP, solubility, and thermal stability using EA, titration, GPC, FTIR and NMR spectroscopy.

Esterification of cellulose continues to provide a dominant route towards cellulose utilization in polymer-based materials. At present homogeneous reaction procedures and *in situ* activation of cellulose with Tos-Cl are increasingly studied since they offer possibilities to novel products with special ester functions of the carboxylic acids. Reactivity and selectivity of the acylation reactions using *in situ* activation with Tos-Cl were studied for different long chain carboxylic acids (capric-, caprylic-, decanoic-, lauric-, palmitic-, stearic acid). Reaction mechanism was studied using ^{1}H NMR spectroscopy. Highly pure products were obtained with high DS of 2.56. Effect of added base pyridine was noted. Products synthesized using pyridine as base showed rather higher DS and DP values comparing with the esters synthesized without pyridine. It is noted that changing molar ratios of reactants and reaction times can control DS of the cellulose esters of long chain fatty acids. Another important finding of this reaction path was that significant DS 1.36 was obtained in only 1 h reaction time at 80°C, which indicates the efficiency of the Tos-Cl towards acylation. The thermogravimetric analysis of these derivatives showed that decomposition temperature (Td) increased with the increase in carbon number starting from 292°C for cellulose caprate to 318°C for cellulose stearate. The esters synthesized were soluble in usual organic solvents depending upon DS.

Novel α-lipoic acid esters of cellulose were homogeneously synthesized with low DS in DMAc/LiCl using differently activated carboxylic acid derivatives. Cellulose α-lipoates were synthesized by the reaction of cellulose with α-lipoic acid after *in situ* activation with Tos-Cl and with novel and efficient reagent for acylation, i.e. CDI. The DS has been determined by mean of EA. The reactions proceeded with high yields. By changing the molar ratio of the reactants, one can control DS. Cellulose α-lipoates prepared with low DS values were soluble in DMSO. Ring of the α-lipoate moiety containing S-S function stays intact during the reaction, which was confirmed by EA, FTIR and ^{1}H NMR spectroscopy. Hence the adsorption of cellulose α-lipoate over gold surface was studied using SPR. Cellulose α-lipoate was used to prepare its thin films over gold surface, which is future aspect of the product to use further in biomineralization.

Besides *in situ* activation of carboxylic acids with CDI, another mild and efficient method is the *in situ* activation of carboxylic acids via iminium chlorides. Iminium chlorides were simply formed by the reaction of DMF with oxalyl chloride. Reaction mechanism of iminium chloride formation was studied using ^{1}H NMR spectroscopy. Esterification of cellulose was carried out using iminium chlorides of carboxylic acids with different substructures, i.e. acetic acid, the long chain aliphatic acids stearic acid and palmitic acid, the aromatic acid 4-nitrobenzoic acid and adamantane 1-carboxylic acid as bulky alicyclic acid. The formation of the iminium chloride and the conversion with the acid were carried out as “one pot reaction”. DS has been determined by means of ^{1}H NMR spectroscopy of completely acylated cellulose samples. Changing the molar ratios of reactants can control DS. The GPC results indicated no significant degradation as DP values 240, 250 and 280 were obtained for different cellulose esters when Avicel® with DP 280 is the starting material. Products obtained were soluble in organic solvents depending on the DS.

Carboxylic acids were efficiently activated with CDI and applied for the acylation of cellulose under homogeneous condition using DMSO/TBAF as solvent system. The simple and elegant method is very mild and easily applicable tool for synthesis of pure aliphatic, alicyclic, bulky and unsaturated carboxylic acid esters of cellulose with DS of 2.2. This mild method showed negligible degradation of cellulose backbone. Besides synthesis of cellulose esters, investigation of reaction mechanism for carboxylic acid imidazolide formation was carried out using ^{1}H NMR spectroscopy. It was found that carboxylic acid imidazolide is the only reactive intermediate under such reaction conditions. Products are soluble in organic solvents, e.g. DMSO or DMF. The cellulose esters prepared were highly pure and showed no impurities or substructures resulting from side reactions. GPC studies revealed fewer degradation of cellulose backbone as DP of 187-228 obtained for cellulose esters prepared by this reaction path.

Novel amphiphilic esters of pullulan with abietic acid were synthesized homogeneously in DMAc/LiCl using differently activated abietic acid. Pullulan abietates were synthesized by the reaction of pullulan with abietic acid after *in situ* activation with Tos-Cl, CDI and iminium chloride. The DS of esters has been determined by means of EA and titration after saponification. All the methods yielded water-soluble products with very low DS required for film formation onto cellulose surface. Pullulan and pullulan abietate showed self-assembly in aqueous media. Adsorption of pullulan and pullulan abietate over the regenerated cellulose surface was studied by SPR, gives further insight into the structure of wood. Pullulan abietate showed more adsorption onto cellulose. GPC studies revealed strong degradation of the pullulan backbone as DP of 32 was observed for pullulan abietate prepared by Tos-Cl, however, DP 95 was obtained for pullulan abietates prepared via iminium chloride method, which appeared less harmful to pullulan backbone. The TGA revealed that thermal decomposition temperature (*T*d) 262°C was obtained for pullulan abietate indicated the polymer obtained is thermally stable.

New hydroxypropyl cellulose (HPC) abietates were synthesized homogeneously by the reaction of HPC with abietic acid after *in situ* activation with Tos-Cl in DMAc. DS were calculated by EA as well as by titration after saponification. The significant DS value 0.91 was obtained for the ester synthesized using 1/1/1 molar ratios of HPC/Tos-Cl/abietic acid. All the products were soluble in usual organic solvents, e.g. DMSO, DMA, $CHCl_3$.

Novel dextran abietates were synthesized homogeneously by the reaction of dextran with abietic acid after *in situ* activation with Tos-Cl in DMAc/LiCl. DS were calculated by EA as well as by titration method. The products with DS of 0.09-0.17 were synthesized, which were soluble in usual organic solvents, e.g. DMSO, DMA, DMF, however, dextran with MW 70,000 yielded water-soluble product with DS_{EA} 0.09.

5. Zusammenfassung

Alternative Wege zur Polysaccharidacylierung: Synthese, Strukturanalytik, Eigenschaften

von M. Sc. M. Phil. Muhammad Ajaz Hussain

1. Einleitung und Aufgabenstellung

Celluloseester sind seit langem bekannte und weitverbreitete semisynthetische Polymere. Beispielsweise wurden Celluloseacetate erstmals 1865 von Schützenberger [1] erwähnt und um die Jahrhundertwende schon industriell produziert. Dabei kamen damals bereits Essigsäureanhydrid als Reagens und Schwefelsäure oder Perchlorsäure als Katalysator zum Einsatz. An dieser Vorgehensweise hat sich prinzipiell bis auf den heutigen Tag praktisch nichts verändert. Zwar ist die Synthese eine Reihe gemischter und komplexer Celluloseester realisiert worden, doch werden dafür meist die Anhydride- oder Chloride der Carbonsäuren umgesetzt. Zur Darstellung maßgeschneiderter Derivate, d.h. Produkte mit speziellen Funktionen und spezieller Verteilung der eingeführten Substituenten, ist es nötig neue Synthesstrategien zu entwickeln.

Neuere Synthesen von Celluloseestern beinhalten vor allem homogene Umsetzungen in Cellulose-Lösemitteln wie *N,N*-Dimethylacetamid (DMA)/LiCl [2], Formaldehyd/ Dimethylsulfoxid (DMSO) [3], Chloral/*N,N*-Dimethylformamid (DMF)/ Pyridin [4] oder *N*-Ethylpyridiniumchlorid-Schmelzen [5-8]. Als Reagenzien kommen neben dem Carbonsäureanhydrid und dem -chlorid auch die Alkali bzw. Erdalkalisalze der Carbonsäure in Kombination mit *p*-Toluolsulfonsäurechlorid [9] zum Einsatz. Modernere Veresterungsverfahren werden unter Einsatz der freien Säure durchgeführt, welche *in situ* in ein reaktives Säurederivat überführt wird. Zur Säureaktivierung wurden N,N-Dicyclohexylcarbodiimide [10,11]; 4-Pyrolidinopyridin [12,13], Methansulfonsäurechlorid bzw. p-Toluonsulfonsäurechlorid (Tosylchlorid) verwendet [14-18]. Es wurden Celluloseester mit langkettigen Fettsäureresten (bis zu C-20, Eikosansäure), ungesättigten (Methacrylat, Zimtsäureester, Vinylessigsäureester) und aromatischen Funktionen dargestellt. Diese Reaktionen sind jedoch mit einer hohen Toxizität des Reagenzes oder zu drastischen Reaktionsbedingungen verknüpft. Polysaccharidester für biotechnologische Anwendungen und Derivate mit sensitiven Strukturen (ungesättigte oder heterocyclische Systeme) sind auf diese Weise nicht zugänglich. Im Rahmen der Arbeit sollten daher effiziente und schonende Wege zur Veresterung von Polysacchariden erforscht werden. Neben der Synthese mussten neue Verfahren zur Strukturaufklärung entwickelt werden, um die unkonventionellen Derivate zu analysieren.

2. Resultate

2.1. Analytik von Celluloseestern

Die sichere und effiziente Analyse des Grades und der Verteilung von Substituenten in der Anhydroglucoseeinheit (AGU) von Celluloseestern ist nach wie vor ein Forschungs- und Entwicklungsanliegen. Die Gründe dafür sind zum einen, dass es durch alternative Veresterungsmethoden möglich war neue Derivate darzustellen, die mit herkömmlichen Mitteln nicht analysiert werden können.

Zum anderen sind detaillierte Struktur-Eigenschafts-Beziehungen, wie die eingeschränkte Löslichkeit von Celluloseacetaten, aufgrund unzureichender Strukturbestimmung noch nicht zugänglich. Neben der Perdeuteroacetylierung von Celluloseacetaten und ^{1}H-NMR spektroskopischer Untersuchung der gemischten Ester, die Goodlett [19] zur Bestimmung der partiellen Substitutionsgrade nutzte, wurden unterschiedliche Verfahren erforscht und hinsichtlich der Reproduzierbarkeit und Aussagekraft beurteilt. Eine geeignete Methode ist die Perpropionylierung der Celluloseester, wobei keine Verunreinigungen des Acylierungsmittels für die Folgeumsetzung mit Essigsäureabkömmlingen stören. Die Zuverlässigkeit dieses Verfahrens wurde sowohl durch Untersuchungen zur Umesterungsneigung der Celluloseacetate bei der Perpropionylierung als auch durch die Bestimmung der Fehlergrenzen der Methode belegt. Eine große Zahl an Celluloseestern mit breiter Varianz an Strukturen konnte analysiert werden. Durch Linienformanalyse der Spektren ist es dabei möglich, neben dem Gesamtsubstitutionsgrad auch die partielle Substitution an den Positionen 2,3 und 6 zu bestimmen.

Da bei ^{1}H-NMR spektroskopischen Untersuchungen an perpropionylierten Derivaten aliphatischer und alicyclischer Celluloseester Signalüberlagerungen die Strukturbestimmung behinderten, wurden zusätzlich Pertrifluoracetylierung und Pernitrobenzoylierung (Bild 1) als Folgereaktionen für die Strukturanalytik mittels ^{1}H-NMR Spektroskopie untersucht. Es zeigte sich, dass die Pernitrobenzoylierung vollständig verläuft und mittels der Methode sicher die Substitutionsgrade von langkettigen aliphatischen Estern wie Stearaten oder Palmitaten bestimmt werden kann. Die Pertrifluoracetylierung ist aufgrund der Hydrolyseinstabilität der Trifluoracetylgruppe nur bedingt einsetzbar.

Eine interessante Alternative ist die Folgederivatisierung mit Isocyanaten. So können Celluloseester mit Ethyl- oder Phenylisocyanat vollständig in die entsprechenden percarbanilierten Produkte überführt und sicher mittels ^{1}H-NMR analysiert werden.

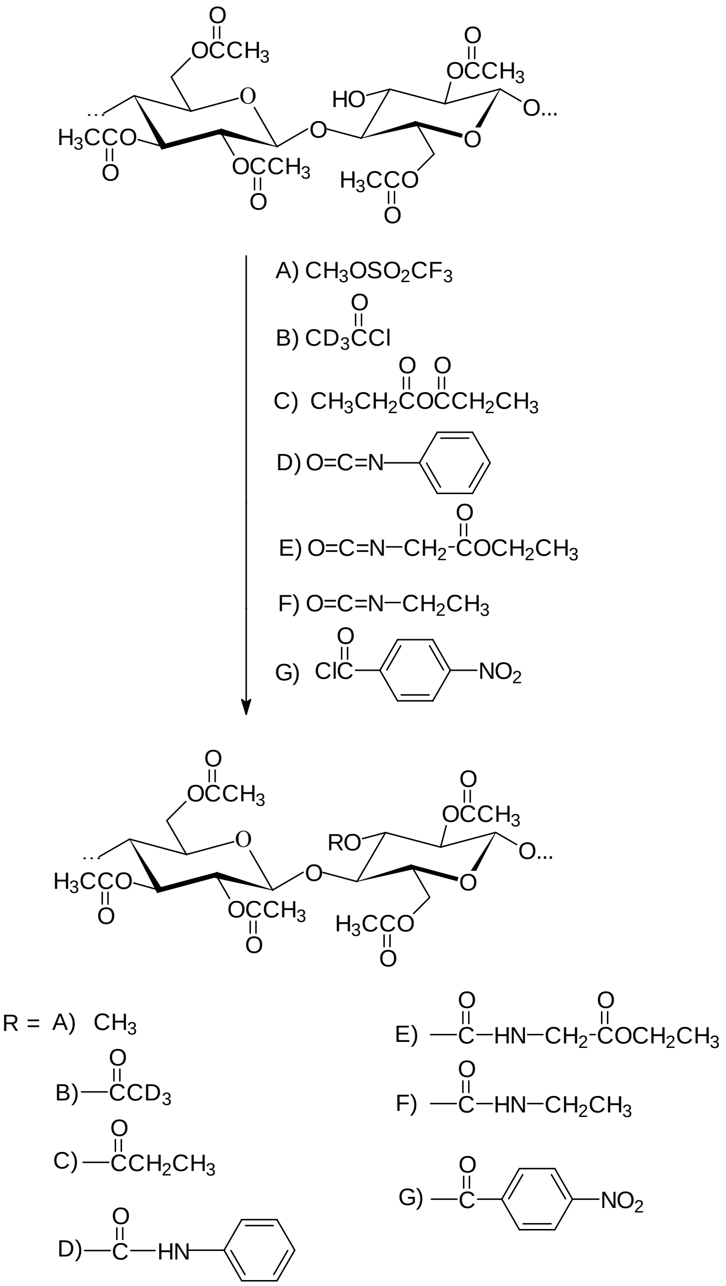


Bild 1: Exemplarische Darstellung der durchgeführten Folgederivatisierung von Celluloseacetat zur Polysaccharidesteranalytik.

2.2. Neue Wege zur Veresterung von Cellulose

2.2.1. In situ *Aktivierung mit p-Toluonsulfonsäurechlorid*

Aufbauend auf Untersuchungen von Koschella et al. [20], Heinze und Schaller [21] und Glasser et al. [22] wurde die Synthese von Celluloseestern unter Verwendung von p-Toluonsulfonsäurechlorid (Tosylchlorid) zur *in situ* Aktivierung der Carbonsäuren durchgeführt. Durch Modellunteruchungen am System Essigsäure/Tosylchlorid mittels ^{1}H-NMR Spektroskopie konnte gezeigt werden, dass das reaktive Intermediat nicht wie aus der Literatur bekannt das gemischte Anhydrid ist, sondern das Carbonsäurechlorid und das symmetrische Anhydrid (Bild 2).

Die Synthese von langkettigen aliphatischen Celluloseestern wurde mittels dieser Strategie studiert. So war die Darstellung von Cellulosecaproaten, -laureaten, -palmitaten und -stearaten mit DS Werten bis 2,56 möglich. Die Produkte konnten mit hoher Reinheit synthetisiert werden und waren entsprechend ihrer DS Werte in diversen organischen Lösungsmitteln wie DMSO, DMF und THF gut löslich. Es wurde der Einsatz von Pyridin als Hilfsbase untersucht, wobei gefunden wurde, dass die Anwendung einer Base bei der Synthese von langkettigen aliphatischen Celluloseestern zu höheren DS Werten als auch zu geringerem Polymerabbau währen der Reaktion führt.

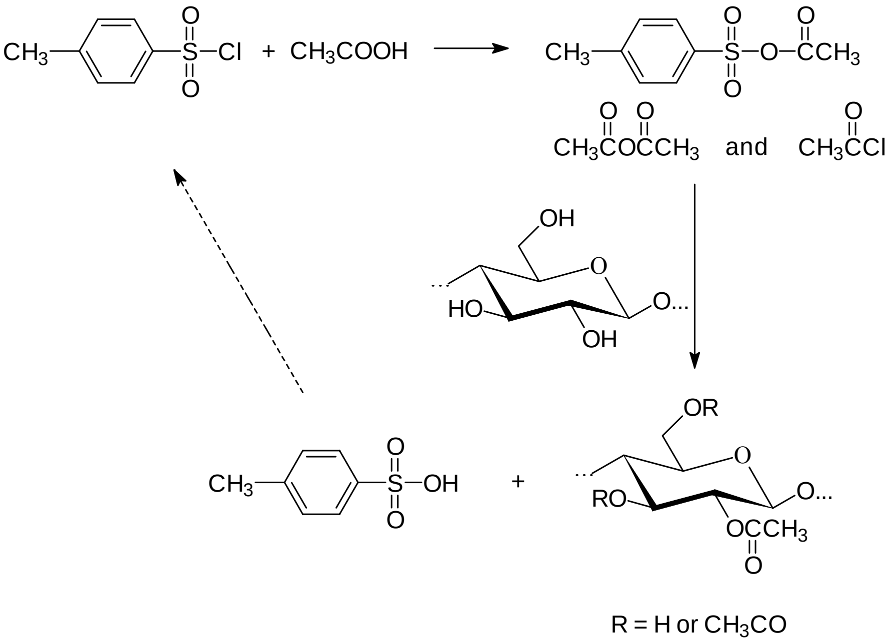


Bild 2: Acylierung von Cellulose nach *in situ* Aktivierung der Carbonsäure (exemplarisch gezeigt für Essigsäure) mit p-Toluonsulfonsäurechlorid.

Neben der Strukturanalyse mittels FTIR, Peracylierung und ^{1}H-NMR Spektroskopie sowie ^{13}C-NMR Spektroskopie wurden die Derivate hinsichtlich ihres thermischen Verhaltens beurteilt. So zeigte die thermogravimetrische Untersuchung der Celluloseester, dass die Zersetzungstemperatur mit der Anzahl der Kohlenstoffatome in der Esterfunktion steigt. Für das Cellulosecaproat wird eine Zersetzungstemperatur von 292°C beobachtet. Im Gegensatz dazu zersetzt sich das Cellulosestearat erst ab 318°C.

2.2.2. In situ *Aktivierung mit N,N-Carbonyldiimidazol (CDI)*

Die Aktivierung der Carbonsäure mit CDI ist für die Derivatisierung von Polysacchariden besonders geeignet, da bei der Umsetzung nur leicht entfernbares Imidazol und CO_2 gebildet werden (Bild 3). Es wurden Modellumsetzungen mit Essigsäure mittels ^{1}H- und ^{13}C-NMR Spektroskopie durchgeführt, um ihre Eignung zur Veresterung von Polysacchariden zu beurteilen. Es konnte gezeigt werden, dass CDI innerhalb von 24 h vollständig zum Säureimidazolid und Imidazol reagiert.

Es wurden verschiedene Ester der Cellulose synthetisiert und charkterisiert. So waren neben Acetaten, Ester der 2-Furancarbonsäure der Adamantancarbonsäure und der α-Liponsäure zugänglich. GPC Untersuchungen belegten, dass die Methode besonders mild abläuft. DP Werte im Bereich von 180-220 wurden bestimmt für Produkte ausgehend von Avicel (DP 280).

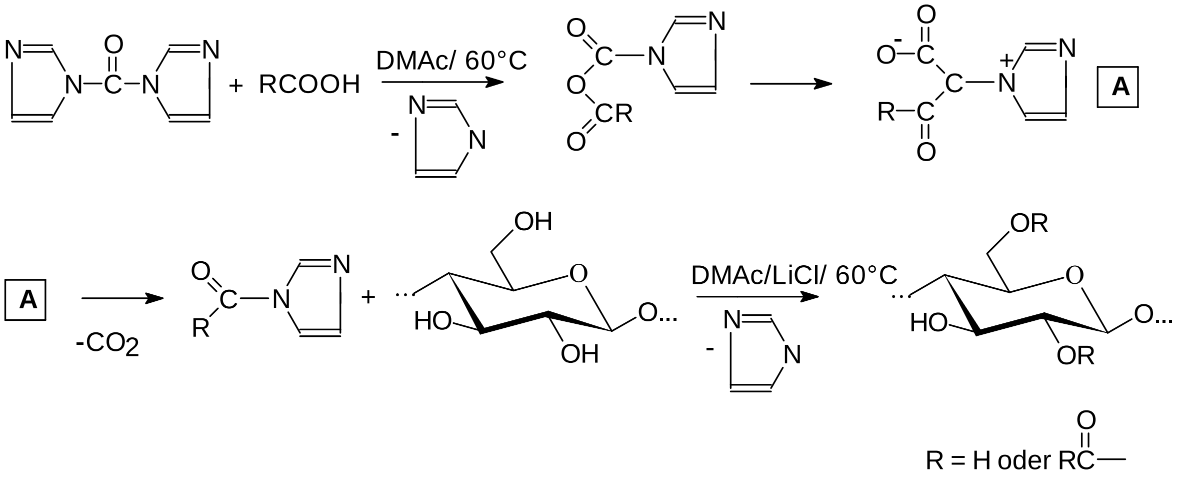


Bild 3: Schematische Darstellung der Celluloseveresterung unter Verwendung von N,N-Carbonyldiimidazol

2.2.3. In situ *Aktivierung mit Imminiumchlorid*

Eine neue und sehr effiziente Methode zur Veresterung von Polysacchariden stellt die „one pot“- Synthese nach *in stiu* Aktivierung der Säure mittels Imminiumchlorid dar, die in der Arbeit erstmals erforscht wurde. Das Reagenz kann in einfacher Weise durch Umsetzung von DMF mit Oxalylchlorid dargestellt werden. Bei der Umsetzung mit der Säure und dem Polysaccharid entstehen nur gasförmige Substanzen und das Lösungsmittel als Nebenprodukte. Dieser Mechanismus wurde mittels ^{1}H-NMR Experimenten belegt.

Die Reaktionsprodukte waren einfach zu isolieren. Der Abbau während der Reaktion ist noch geringer als im Falle der Umsetzung mit CDI. So wurden bei Synthesen ausgehend von Avicel (DP 280) Produkte mit DP Werten von 220-280. Auch dieses Verfahren ist für die Synthese verschiedenster Estertypen genutzt worden. So waren auf diesem simplen Weg nebenproduktfreie Celluloseacetate, -nitobenzoate, -adamantate, -palmitate und -stearate herstellbar. Die höchsten DS Werte werden für die aliphatischen Säuren erhalten.

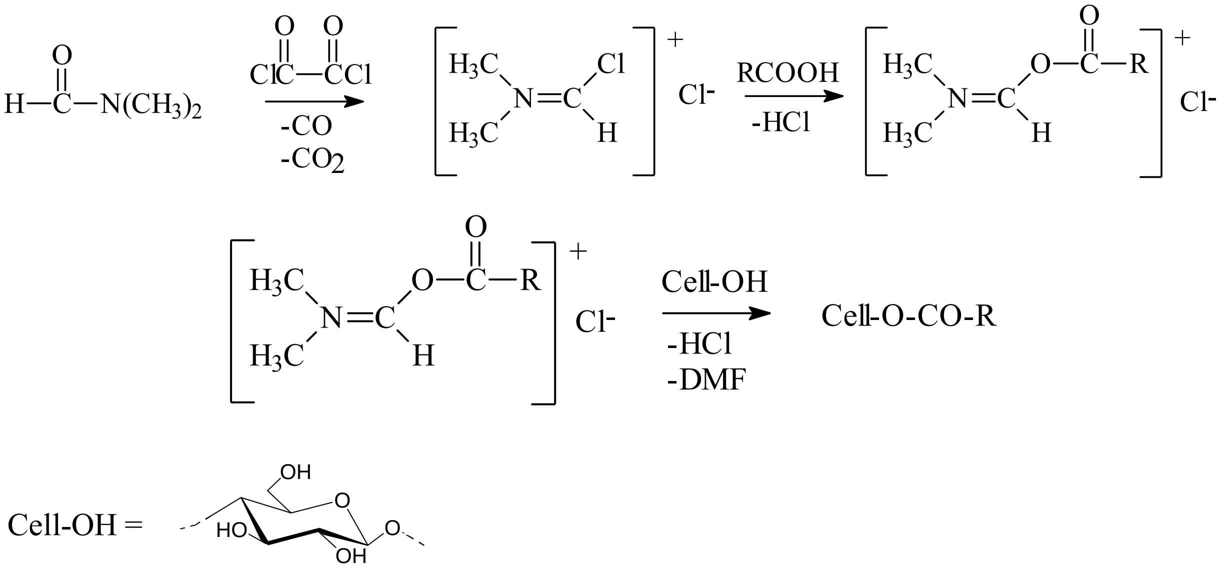


Bild 4: Schematische Darstellung der Celluloseveresterung unter Verwendung von Imminiumchlorid.

2.2.4. Veresterung im neuen Cellulose-Lösemittel DMSO/TBAF

Ein einfach zu handhabendes, unkonventionelles Lösungsmittel für Cellulose ist das Gemisch DMSO/Tetrabutylammoniumfluoridtrihydrat (TBAF). Cellulose mit einem DP Werten bis 650 wird darin innerhalb von 15 min klar gelöst. Da bekannt war, dass die Verwendung von

CDI zur Veresterung auch für wasserhaltige Reaktionsmedien möglich ist, wurde die Derivatisierung von Cellulose in diesem Lösemittel erforscht. Diese Vorgehensweise ist eine der einfachsten und schnellsten Synthesevarianten zur Darstellung reiner, hochsubstituierter, hochmolekularer Celluloseester mit einer breiten Varianz an Substrukturen. Bemerkenswert war das Celluloseester mit sehr hohen Reagenzausbeuten dargestellt werden konnten. So war die Synthese eine Cellulose-2-furancarbonsäureesters (DS 1,91) mit einer Effizienz von 61 % möglich.

2.3. Neue Polysaccharidester

Neben Celluloseestern wie Celluloseacetate, -nitobenzoate, -adamantate, -furoaten, -palmitate und -stearate wurden auch neue Derivate synthetisiert und charakterisiert. So wird erstmals die Darstellung eines löslichen Cellulose-α-liponsäureesters realisiert. Die Ester zeigen aufgrund ihrer S-S Funktion im Substituenten eine gute Absorption an Gold-Oberflächen was mittels Surface Plasmon Resonance (SPR) Spektroskopie untersucht wurde. Es werden Schichtdicken zwischen 9 – 49 Å gefunden.

Neben der Funktionalisierung von Cellulose wurden die neu entwickelten Methoden auch zur Derivatisierung anderer Polysaccharide eingesetzt. So konnten Pullulan- und Dextranabietate mit definierter Löslichkeit dargestellt werden, die hinsichtlich ihrer Wechselwirkung mit Celluloseschichten erforscht wurden um die Möglichkeit der Entwicklung von Biomemetika für Holz zu studieren. Mittels Atomkraftmikroskopie und SPR Spektroskopie konnte gezeigt werden, dass niedrig substituierte Pullulanabietate auf Cellulosemonoschichten absorbiert werden.

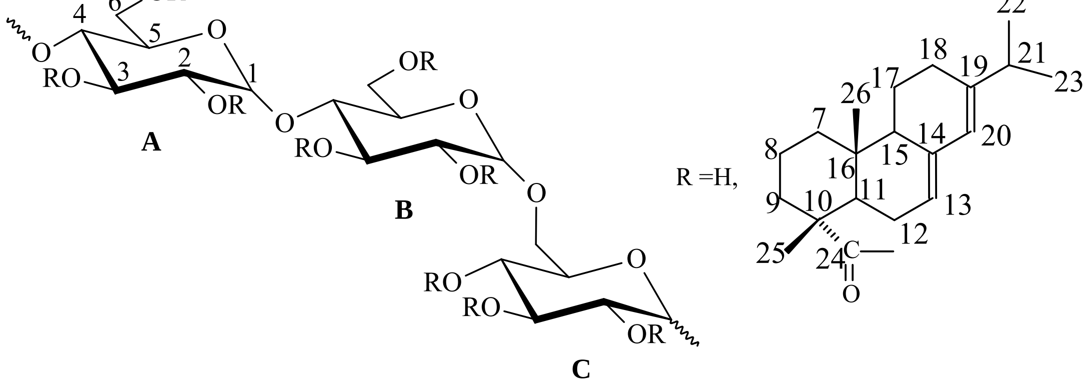


Bild 5: Struktur eines erstmals synthetisierten Pullulanabietats.

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Selbständigkeitserklärung

Ich erkläre, dass ich die vorliegende Arbeit selbständig und unter Verwendung der angegebenen Hilfsmittel, persönlichen Mitteilungen und Quellen angefertigt habe.

Jena, den 06.02.2004 --

Unterschrift