Advanced Solid State NMR Spectroscopic Techniques
in the Study of Thermally Modified Wood

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ABSTRACT

Wood is thermally modified to obtain desired properties such as dimensional stability, low equilibrium moisture content and increased resistance to biodegradation and weather. Our detailed understanding of the changes in the chemical structure of wood induced by thermal treatment is not entirely complete. Furthermore, the effects of thermal modification on the chemistry of wood biodegradation and weathering are not well known despite the widespread use of thermally modified wood as exterior material. Various solid state ^{13}C CPMAS NMR measurements were applied to investigate the changes in chemical structure induced by a Finnish industrial-scale heat treatment process. Several wood species, including pine, spruce, birch, aspen and oak were studied. Wood samples were thermally modified under normal pressure using water vapour as a shielding gas, in the temperature range of 160-240°C. Some of the thermally modified and unmodified wood samples were further exposed to soft or brown rot fungi and weathering. Besides conventional ^{13}C CPMAS NMR measurements, advanced solid state NMR techniques, i.e., spin-locking and dipolar dephasing, were applied to investigate the changes in the main components of wood: cellulose, hemicelluloses and lignin. The effect on wood of this type of thermal modification has not previously been studied by advanced solid state NMR techniques.

According to conventional ^{13}C CPMAS NMR spectra, degradation and deacetylation of hemicelluloses occurred during thermal modification of both softwoods and hardwoods. However, overlapping of the signals of different wood components makes the investigation of the cellulose content difficult. The subspectra of cellulose and the lignin-hemicellulose matrix were separated by the spin-locking technique and linear combination. The results showed the cellulose crystallinity index (CrI) to increase in thermal modification for every wood sample, as a consequence of the preferred degradation of the less ordered carbohydrates. Both before and after thermal modification, the CrI values were higher for softwoods than hardwoods: values for softwood samples ranged from 51% before to 65% after, and values for hardwood samples from 41% before to 54% afterwards. Sample pairs were taken from the same tree trunk in order to eliminate unnecessary errors due to heterogeneity of the wood structure. Differences in the CrI values of samples taken from the same trunk were found to be minor. The accuracy of the spin-locking technique was evaluated by comparing cellulose crystallinity values obtained by the spin-locking technique and the wood crystallinity values obtained by X-ray scattering measurements for the same set of pine and spruce wood samples. The values were in good agreement.

Dipolar dephasing technique was applied to study the lignin quaternary carbons and the degree of condensation. Besides the cleavage of the β-O-4 linkages and demethoxylation in both hardwood and softwood lignin, condensation was observed in softwood lignin after the thermal modification.

^{13}C CPMAS NMR spectral data were interpreted by principal component analysis (PCA) to study thermally modified and unmodified pine wood samples that had been exposed to either soft or brown rot fungi. With PCA wood samples could be classified according to their weight losses and heat treatment temperatures. Weight losses were largest for the unmodified wood samples (varying from 40% to 60%) and least for the samples heat treated at over 220°C (~2-3%). Exposure of unmodified pine to fungi resulted in a drastic decay of the cell wall polysaccharides. Soft rot fungus attacked cellulose more extensively, while brown rot fungus degraded mainly hemicelluloses. The changes in the lignin structure were minor. Increased biological resistance of thermally modified pine was observed.

Conventional ^{13}C CPMAS with dipolar dephasing was applied to examine thermally modified and unmodified pine wood samples that had been exposed to natural weathering for seven years. The NMR spectral data revealed a significant decrease in lignin content of the weathered thermally modified and especially the weathered unmodified samples. According to the results, the surface of the thermally modified sample was still rich in aromatic and conjugated carbonyl structures, whereas the surface of the unmodified sample was enriched in cellulose. The structure of the thermally modified wood sample was modified and the degradation products were not leached out with water as easily as from the unmodified sample. Increased weather resistance was observed for thermally modified pine.

PREFACE

Helsinki, December 2004

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ABBREVIATIONS AND SYMBOLS

am	amorphous
CPMAS	cross polarisation magic angle spinning
cr	crystalline
DD	dipolar dephasing
e	etherified C-4
f	free phenolic C-4
FT-IR	Fourier Transform Infrared
G	guaiacyl
GC-MS	GasChromatography Mass Spectrometry
h	hemicelluloses
MWL	milled wood lignin
NMR	nuclear magnetic resonance
p[1]	first principal component
p[2]	second principal component
PCA	principal component analysis
PSRE	proton spin-relaxation based spectral edition
RASH	rapid steam hydrolysis
ref	reference
S	syringyl
s	substituted
SR	soft rotted
tm	thermally modified
UV	ultraviolet
UV-VIS	ultraviolet-visible light
WAXS	wide angle X-ray scattering
wl	weight loss

CrI	crystallinity index
d2	dipolar dephasing delay
T_1	spin-lattice relaxation time
T_2	spin-spin relaxation time
T_{1H}	proton spin-lattice relaxation time
$T_{1\rho H}$	proton spin-lattice relaxation time in rotating frame
t_a	acquisition time
t_{cntct}	contact time
t_{sl}	spin-lock delay
t_{90}	90° proton preparation pulse

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PREFACE

ABBREVIATIONS AND SYMBOLS

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LIST OF ORIGINAL PUBLICATIONS

AUTHOR´S CONTRIBUTION TO THE PUBLICATIONS

LIST OF ORIGINAL PUBLICATIONS

This thesis is based on the following five publications, which are referred to in the text by their Roman numerals I-V:

I   Sivonen, H., Maunu, S.L., Sundholm, F., Jämsä, S. and Viitaniemi, P., Magnetic resonance studies of thermally modified wood, *Holzforschung* **56** (2002) 648-654.

II   Sivonen, H., Nuopponen, M., Maunu, S.L., Sundholm, F. and Vuorinen, T., Carbon-thirteen cross-polarization magic angle spinning nuclear magnetic resonance and fourier transform infrared studies of thermally modified wood exposed to brown and soft rot fungi, *Appl. Spectrosc.* **57** (2003) 266-273.

III   Nuopponen, M., Wikberg, H., Vuorinen, T., Maunu, S.L., Jämsä, S. and Viitaniemi, P., Heat-treated softwood exposed to weathering, *J. Appl. Polym. Sci.* **91** (2004) 2128-2134.

IV   Andersson, S., Wikberg, H., Pesonen, E., Maunu, S.L. and Serimaa, R., Studies of crystallinity of Scots pine and Norway spruce cellulose, *Trees* **18** (2004) 346-353.

V   Wikberg, H. and Maunu, S.L., Characterisation of thermally modified hard- and softwoods by ^{13}C CPMAS NMR, *Carbohydr. Polym.* **58** (2004) 461-466.

AUTHOR'S CONTRIBUTION TO THE PUBLICATIONS

Publication I

Hanne Wikberg (née Sivonen) drew up the research plan and wrote the manuscript. Hanne Wikberg was responsible for the NMR spectroscopic measurements and Saila Jämsä for the ESR spectroscopic measurements.

Publications II and III

Hanne Wikberg (née Sivonen) drew up the research plan and wrote the manuscripts in close co-operation with Mari Nuopponen. Hanne Wikberg was responsible for the NMR spectroscopic measurements and Mari Nuopponen for the FT-IR and UVRR spectroscopic measurements.

Publication IV

Hanne Wikberg drew up the research plan and wrote the manuscripts in close co-operation with Seppo Andersson. Hanne Wikberg was responsible for the NMR spectroscopic measurements and Seppo Andersson for the X-ray scattering measurements.

Publication V

Hanne Wikberg drew up the research plan and wrote the manuscript.

1 INTRODUCTION

In many applications, wood is used native. There is also a need, however, for modified wood, with properties tailored for specific purposes. Resistance to biodegradation and weathering is a frequently desired property today. For several decades now, thermal treatment has been used to modify the properties of wood. Many different methods have been reported, the first of them dating back to the 1920s. In the intervening years, methods have been much improved, and the applications of wood have diversified considerably as a result. Work on thermal modification processes for timber has currently being done for example in France, the Netherlands and Finland. The product developed in France is called torrified wood.[1-5] The process relies on the use of nitrogen as a shielding gas at atmospheric pressure. In the Netherlands, plato wood is a product of a two-step process where the wood is heated at 150-180°C in aqueous environment at superatmospheric pressure.[6] The process developed at the Technical Research Centre of Finland, VTT, together with Finnish industry is based on heating wood at temperatures of 180-250°C, at atmospheric pressure, and using water vapour as a shielding gas.[7,8] The product is called ThermoWood. The objective of the present work was, through study of thermally modified and unmodified wood by various solid state ^{13}C CPMAS NMR spectroscopic techniques, to clarify the changes in chemical structure induced by this Finnish industrial-scale heat treatment process. Part of the thermally modified and unmodified wood samples were exposed to fungi and weathering.

The chemical modifications in wood structure occurring at high temperature are accompanied by several favourable changes in the physical structure: reduced shrinkage and swelling, low equilibrium moisture content, increased thermal insulating capacity, better decay resistance, enhanced weather resistance and dark, decorative colour.[1,3,8-14] All these changes in structure are achieved without addition of chemicals. Thermal modification can thus be considered as a wood preservative treatment without addition of harmful chemicals, producing a durable material with desired properties and appearance. Thermally modified wood is considered an ecological alternative to impregnated wood.[1]

Industrial output of thermally modified timber in Finland was about 25 000 m^3 in the year 2003.[15] The amount is expected to increase in future with the appearance of several new companies and growing demand. Thermally modified wood finds use in many outdoor and indoor applications; in garden, kitchen, bathroom and sauna furniture; in floors, ceilings, doors and window joinery; in musical instruments; and in a diversity of other outdoor and indoor applications. Unfortunately, the mechanical strength of wood is reduced in the process. Viitaniemi and Jämsä[8] and Viitaniemi[10] have reported that the bending strength of thermally modified wood is reduced by 0-30% depending on the treatment conditions. An average decrease in bending strength as high as 44-50% has also been reported.[16] Thermally modified wood is not recommended, therefore, for constructions under load.

2 CHEMICAL COMPOSITION OF WOOD

Wood is a complex composite material, which consists mainly of cellulose (40-50% of the dry wood), hemicelluloses (25-35% of the dry wood), lignin (20-30% of the dry wood) and low-molecular-mass compounds, extractives (< 5% of the dry wood).[17,18] In the wood cell walls, the cellulose microfibrils are oriented axially giving wood its strength. Hemicelluloses and lignin act as solidifying agents between these cellulose microfibrils. Knowledge of the chemical structure of wood constituents is a prerequisite for understanding the numerous changes in wood that occur during heating.

2.1 Cellulose

Cellulose is the main polymeric component of the wood cell wall, consisting of anhydro-β-D-glucopyranose units linked by (1→4)-glycosidic bonds to form linear high-molecular-mass polymeric chains of over 10 000 glucose residues (Fig. 2-1).[18]
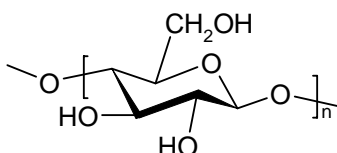
Figure 2-1. Molecular structure of cellulose chain.[17,18]

The hydroxyl groups in cellulose form intra- and intermolecular hydrogen bonds. Bundles of cellulose molecules are thus aggregated together forming partially crystalline microfibrils that are surrounded by amorphous hemicelluloses and lignin. The cellulose microfibrils are oriented in different directions in the different fibre wall layers, and they contain crystalline regions. In addition, their orientation (i.e. a specific microfibril angle) has an influence on the physical properties of a wood fibre. Microfibrils are combined to greater fibrils and lamellæ.

2.2 Hemicelluloses

Hemicelluloses are branched and amorphous heteropolysaccharides, which consist of different monosaccharide units together with some moieties of deoxyhexoses and hexauronic acids. The main constituents of the hemicelluloses are five neutral sugars: glucose, mannose, galactose, xylose and arabinose.[17,18] Hardwoods contain more hemicelluloses than softwoods and the composition is different.

2.2.1 Softwoods

Galactoglucomannans (about 20%) are the main hemicelluloses in softwoods (Fig. 2-2).[18] In addition, softwoods contain arabinoglucuronoxylan (5-10%), arabinogalactan and other polysaccharides.
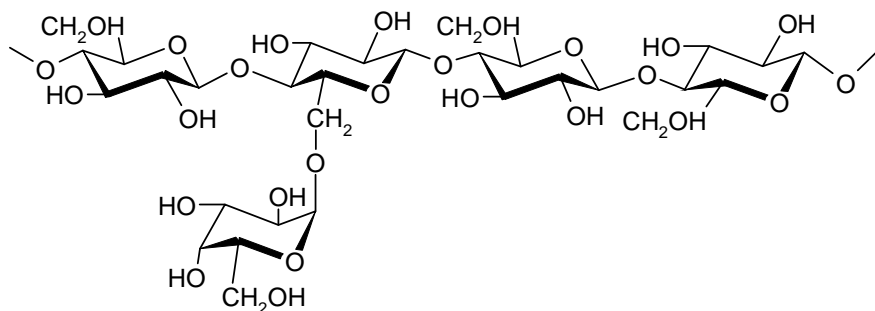
Figure 2-2. *Principal chemical structure of galactoglucomannans.*[17,18]

2.2.2 Hardwoods

Glucuronoxylans are the main hemicelluloses in hardwoods (Fig. 2-3). Depending on the hardwood species, xylan content varies between 15 and 30% of the dry wood.[18] Besides xylan, hardwoods contain glucomannan (2-5%) and other polysaccharides.
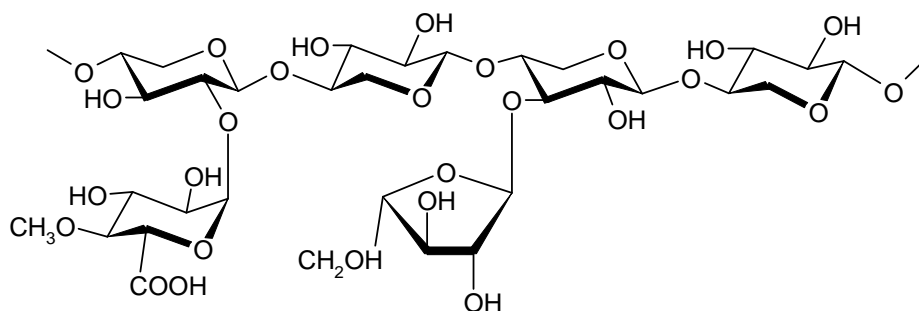
Figure 2-3. *Principal chemical structure of arabinoglucuronoxylan.*[17,18]

2.3 Lignin

Lignin is the second most abundant biopolymer in the plant world after cellulose. Lignins are complex heterogeneous polymers of phenylpropane units, or more precisely polymers of *trans*-coniferyl, *trans*-sinapyl and *trans*-*p*-coumaryl alcohols, which are called the precursors of lignin (Fig. 2-4). These phenylpropane units are joined together by ether or carbon-carbon

linkages. Approximately fifty to sixty per cent of all linkages in lignin are β-O-4 aryl ether linkages.[17,18]
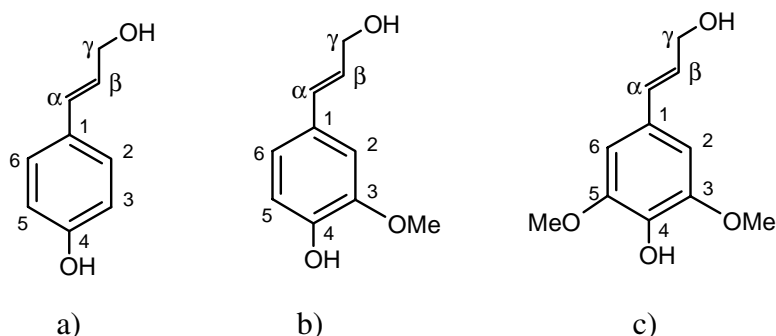
Figure 2-4. *The monomeric units in lignin: a) trans-p-coumaryl alcohol, b) trans-coniferyl alcohol (guaiacyl unit) and c) trans-sinapyl alcohol (syringyl unit).*[17,18]

The lignin in softwoods is mainly composed of guaiacyl (G) units. The lignin in hardwoods is built up of G and syringyl (S) units in ratio varying from 4:1 to 1:2 for the two monomeric units.[18]

2.4 Extractives

Though the low-molecular-mass substances comprise only a few per cent of the dry wood mass, they have a significant influence on the properties of wood. Extractives are compounds that can be extracted from wood with polar or non-polar solvents. Many organic compounds are included among extractives, e.g., terpenes and terpenoids, esters of fatty acids (fats and waxes), phenolic compounds and several other compounds. Extractives are the non-structural compounds of wood and they are not a part of this thesis.

3 NMR STUDIES OF LIGNOCELLULOSICS AFTER DIFFERENT EXPOSURES

The complex structure of wood sets conditions for measuring equipment. Nuclear magnetic resonance (NMR) spectroscopy is a proven tool in the analysis of wood and its components. Model compounds and isolated preparations of wood components are accurately characterised by several liquid state NMR techniques, while the structure of native wood has been less often characterised. Unfortunately, the necessary step of isolation or fractionation may cause significant modification in the structure of wood. An essential advantage of solid state NMR spectroscopy is that the samples can be studied in native state without component isolation or fractionation, and all chemical changes in the structure that may occur in chemical treatment are thereby avoided.

3.1 Heat and steam

^{13}C CPMAS NMR has been applied to analyse the chemical structure of various thermally modified wood species, including pine, spruce, beech, oak, aspen and poplar. According to published results, the main effects of thermal modification on the chemical structure of wood are degradation of hemicelluloses and amorphous cellulose and demethoxylation and depolymerisation of lignin. Some variations are reported depending on the heat treatment process and the wood species.

Tjeerdsma et al.[6] applied solid state NMR to study the chemical changes induced by mild heat treatment in two steps in common beech (*Fagus sylvatica*) and Scots pine *(Pinus sylvestris)*. The content of amorphous carbohydrates was decreased during treatment and a variety of minor changes occurred in the lignin structure, including lignin depolymerisation and demethoxylation. Relaxation measurements have been applied extensively to study the interactions between wood components. Košiková et al.,[19] for example, investigated structural modifications and the interaction of lignin and polysaccharides in common beech *(Fagus sylvatica)* during drying, using ^{13}C CPMAS NMR analysis together with relaxation ($T_{1\rho H}$) measurements. A comparison of the CPMAS spectra of untreated dried wood and wood that had been pretreated by steam or NaOH or both and then dried revealed similar results to those obtained by others. The uniform relaxation times of lignin and cellulose were used to confirm the interaction between these components. Relaxation measurements have also been applied to study free radicals in heat treated wood. Baldock and Smernik[20] heat treated pine (*Pinus resinosa*) at different temperatures up to 350°C. Shorter NMR relaxation times (T_{1H} and $T_{1\rho H}$) with higher treatment temperature were applied to confirm the increase in the concentration of unpaired electrons (free radicals) in the heated wood samples. Recently, Pétrissans et al.[4] studied the wettability and chemical composition of heat treated Norway spruce (*Picea abies*), Scots pine (*Pinus sylvestris*), black poplar (*Populus nigra*) and

common beech (*Fagus sylvatica*) by CPMAS together with FT-IR. On the assumption that amorphous cellulose absorbs more water than crystalline cellulose, they suggest that the increase they found in cellulose crystallinity may be the reason for the more hydrophobic nature of heat treated wood. Unfortunately, only the CPMAS spectra of spruce were published, and differences in the behaviour of wood species were not discussed.

Steam is used as a shielding gas in the heat treatment process developed at VTT. Several NMR studies in liquid[21] and in solid state[22-25] on the effect of steam in steam explosion processes of wood have been reported. Steam explosion is normally used to separate lignocellulosic material into its main components. In the process, wood chips or shavings are treated with pressurised steam, which is then quickly released through a valve. After a steam explosion, about 50% of the wood is obtained as a solid residue, mostly consisting of carbohydrates, but also some lignin. Hydrolysis of hemicelluloses and cleavage of β-aryl ether linkages in lignin are reported.

Hemmingson and Newman,[23] using ^{13}C CPMAS NMR, examined Monterey pine (*Pinus radiata*) and mountain ash (*Eucalyptus regnans*) before and after two different steam explosion processes. The main structural effects were similar for the two species, though some differences were detected in the cellulose crystallinity and the structure of residual lignin. Tekely and Vignon[24] applied relaxation measurements to analyse the mobility of different components in a solid matrix. Relaxation times of lignin and cellulose in common aspen (*Populus tremula*) were found to be of different magnitude after steam explosion. Two possible explanations were presented: the recondensation of lignin during steam explosion leading to a change in molecular mobility, or the removal of either noncrystalline cellulose or hemicelluloses leading to a more homogeneous polymer. Josefsson et al.[25] analysed the cellulose supramolecular structure in pulps obtained by steam explosion of common aspen (*Populus tremula*) by ^{13}C CPMAS NMR together with chemometrics. Interpretation of NMR spectral data by principal component analysis (PCA) was used to categorise the changes in the cellulose supramolecular structure as two independent processes, which were discussed in detail.

Rapid steam hydrolysis (RASH) of a red oak (*Quercus rubra*) has been investigated by ^{13}C CPMAS NMR.[22] RASH is similar to steam explosion, except that no sudden decompression is applied and steam is continuously introduced during the treatment, with volatile and water-soluble products (principally depolymerised hemicelluloses and lignin) continuously removed from the reactor and recovered with the steam condensate. In this process, wood chips of red oak were subjected to high pressure and temperatures in the range of 200 to 280°C. The cleavage of aryl ether linkages was most extensive for the sample treated at highest temperature.

3.2 Biodegradation

Enhancing the resistance of wood to biodegradation in order to maintain the desired properties during ground contact is an important goal of timber technology. Thermal modification of wood has been found to improve bioresistance.[1,3,8-10] Pine and poplar treated under relatively mild thermal conditions have recently been investigated by gas chromatography-mass spectrometry (GC-MS) and by liquid state ^{13}C NMR spectroscopy.[1] The formation of small amounts of some polynuclear aromatic hydrocarbon derivatives of phenanthrene, as well as other classes of polyaromatic compounds, was observed. The authors suggest that the fungal resistance of thermally modified wood can be attributed, at least in part, to the polyaromatics formed from extractives, lignin, hemicelluloses and cellulose during thermal treatment. Wood extractives have long been recognised as a probable source of the natural resistance of certain wood species against fungal attack.[17,18] The location of the extractives also plays an important role. Thus heartwood, which contains more secondary extractives (e.g. phenolic compounds), is more resistant than sapwood.[26-29] However, at least some extractives, such as fatty acids, fats and waxes, migrate to the surface of heat-treated wood, and treatment at high temperatures removes them from the wood.[30]

Weiland and Guyonnet[3] offer some possible explanations for the improved bioresistance of thermally modified wood: 1) new thermal modification products arising from the treatment, which act as fungicides, 2) new modified wood components, which the fungi no longer recognise and are therefore incapable of degrading and 3) elimination of hemicelluloses, which then no longer act as a nutritive for fungi. The cause of the durability of thermally modified wood against fungi appears to be complex, however, and probably more than one factor is involved.

Better understanding of the improved bioresistance of thermally modified wood requires an examination of the chemistry behind wood biodegradation. Biodegradation pathways in wood depend on several factors, such as the particular fungus and wood species. Most white rot fungi attack hardwoods, while most brown rot fungi attack softwoods.[17] White rot fungi are capable of decomposing all wood components, whereas brown rot fungi primarily decompose the carbohydrate fraction of wood. When wood is exposed to brown rot fungi, it rapidly loses its mechanical strength and undergoes drastic shrinkage and cracking across the decayed wood. After a certain time, the residual wood is mainly composed of structurally modified lignin.[31] The fungi causing soft rot attack wood exposed to moisture and remove significant amounts of carbohydrates from the cell wall. Although the degradation pathway depends on the species, soft rot fungi can degrade both softwood and hardwood.[17] As a result of soft rot degradation, the strength of wood is reduced.[31]

NMR, both in solid and in liquid state, has been applied to investigate the chemical changes in wood due to fungi attack. Solid state NMR studies on wood exposed to brown and white rot fungi have appeared, but no NMR studies on soft rot fungi. The main chemical changes

in wood due to the biodegrading activity of brown and soft rot fungi are loss of hemicelluloses and amorphous cellulose. Some changes have been observed in the structure of lignin as well. Cleavage of β-O-4 linkages and Cα-Cβ bond cleavage as well as loss of methoxyl groups are reported.

Measurements by ^{13}C CPMAS NMR with signal integration of decayed and undecayed wood have been used to study the biodegradation of several wood species.[32-35] Davis et al.[36-38] applied CPMAS and DD measurements in combination with peak deconvolution and spectra subtractions to investigate the decay of paper birch (*Betula papyrifera*) and Colorado spruce (*Picea pungens*) due to several white rot fungi and brown rot fungus. Kim and Newman[39] studied brown rot decay of Korean pine (*Pinus koraiensis*) using proton spin relaxation editing (PSRE) to separate the subspectra of cellulose and noncellulosic material, and then further resolution enhancement to study the crystalline forms of cellulose. Gilardi et al.[40] used CPMAS and DD measurements and peak deconvolution combined with FT-IR measurements to analyse Scots pine (*Pinus sylvestris*) and common beech (*Fagus sylvatica*) wood cell wall biodegradation by brown and white rot fungi. Conclusions were also drawn from NMR spectral features such as line widths, which gave information about modification in the mobility of molecular fragments. Sharpening of lignin resonances after the attack was considered as a sign of higher mobility of the fragments.

Wide-line solid state NMR method based on T_1 and T_2 relaxation times has been used to analyse common beech (*Fagus sylvatica*) and Scots pine (*Pinus sylvestris*) samples subjected to fungal attack.[41] Increase in the relaxation times of decayed relative to undecayed wood samples was attributed to an increase in the mobility of the molecular components of the cell wall and of the water in the cell walls, as a consequence of the decay by fungal enzymes.

3.3   Weathering

The effects of thermal modification on the chemistry of wood weathering are not well known despite the use of thermally modified wood as exterior construction material, where weather resistance is essential. The need for the characteristics and appearance of wood to be maintained, while it is exposed to sunshine and rainfall is assuming increasing importance as the outdoor uses of wood-based materials expand. Unprotected wood exposed to outdoor conditions undergoes a variety of degradation reactions induced by factors as diverse as light, moisture, heat, oxygen and pollutants.[42] The weathering of wood is primarily a surface phenomenon, although the cracks developing during weathering will be sensitive to further attack by fungi and lead to more severe destruction.

UV light is fundamentally involved in the degradation and discoloration of wood surfaces in natural weathering, where it causes depolymerisation of lignin in the wood cell wall; water then erodes the surface by washing away the degradation products. The main UV light-

absorbing component of wood is lignin. Pure cellulose and hemicelluloses absorb little UV light and usually it is the presence of lignin that causes the photodegradation of carbohydrates. In addition, some of the unsaturated wood extractives, such as resin acids in softwoods, may participate in light-induced free-radical reactions.[42] Most of the free radicals in wood are generated in phenolic groups of lignin, which are then easily converted to quinonoid structures. Progressive destruction of the aromaticity of lignin and the formation of carbonyl structures are well-known changes due to weathering. The relative content of cellulose has also been reported to increase because of the degradation and solubilisation of lignin.[42] After the photochemical and photophysical reactions in wood components, the characteristics and properties of the wood are changed.

Few studies on the weathering of lignocellulosics by NMR have been published. Hemmingson and Wong[43,44] and Hemmingson and Morgan[45] studied the photodegradation process in newsprint exposed to direct or indirect sunlight by ^{13}C CPMAS and DD as well as by liquid state NMR. Progressive destruction of lignin aromaticity, demethoxylation and formation of carbonyl groups were detected. Degradation also produced soluble carbohydrate and lignin fragments. The acetone-water solubles were complex mixtures of lignin fragments as well as resin and fatty acid extractives. Oxidative cleavage of the Cα-Cβ bonds was the predominant degradation pathway conserving the aromaticity of lignin fragments.

Light-induced degradation of sheets of spruce milled wood lignin (MWL) has been investigated by liquid state ^{1}H and ^{13}C NMR after lignin was extracted from the irradiated sheets.[46] α- and β-ether and α-β bond cleavage in lignin were the main reactions, while the main degradation products were vanillin and vanillic acid and the amount of aromatic aldehyde and carboxyl groups was increased. Kimura et al.[47] used DD technique and spectra subtraction to study lignin photodegradation in unbleached and alkaline hydrogen peroxide bleached black spruce stoneground wood pulps, and they observed that both the etherified and nonetherified guaiacyl structures contribute to the significant loss of lignin aromaticity. The light stability of wood panel treated with different anhydrides has recently been examined.[48] ^{13}C CPMAS NMR was applied to investigate the structure of acetylated wood and it was observed that esterification of the holocelluloses occurred mainly at the amorphous C-6 position. It was also observed, with use of UV-VIS spectrophotometry, that the colour difference and yellowness index of acetylated wood after irradiation were significantly less than in controls, indicating that acetylation inhibits the photoyellowing of wood.

4 OBJECTIVES OF THE STUDY

Our detailed understanding of the changes that thermal modification causes in the chemical structure of wood is not entirely complete. Furthermore, the effects of thermal modification on the chemistry of wood biodegradation and weathering are not well known despite the widespread use of thermally modified wood as an exterior material where bio- and weather resistance are essential. Understanding the changes resulting from the thermal modification of wood requires detailed study of the chemical structure. The objective of the present work was, through study of thermally modified and unmodified wood by various solid state ^{13}C CPMAS NMR spectroscopic techniques, to clarify the changes in chemical structure induced by the Finnish industrial-scale heat treatment process. Part of the thermally modified and unmodified wood samples were exposed to fungi and weathering. The effects of this type of thermal modification have not been studied previously by advanced solid state NMR techniques.

Solid state NMR spectroscopy is a highly useful tool in the analysis of wood due to its nondestructive nature: the wood samples can be studied in native state. Evaluation of advanced solid state NMR techniques (e.g., spin-locking and dipolar dephasing techniques) for the study of wood components cellulose, hemicelluloses and lignin in pine, spruce, birch, aspen and oak, before and after thermal modification, was a major goal of the work.

5 MATERIALS

5.1 Thermal modification process and timber

The thermal modification process for wood developed at the Technical Research Centre of Finland, VTT, is carried out under atmospheric pressure, with water vapour as a shielding gas.[8] Wood is heated for several hours at high temperatures in the range of 180-250°C. The process can be tailored for a particular end use by optimising the temperature and duration of treatment.

Two softwoods and three hardwoods were studied: Scots pine (*Pinus sylvestris*), Norway spruce (*Picea abies*), silver birch (*Betula pendula*), European aspen (*Populus tremula*) and European oak (*Quercus robur*). Wood boards were divided into two pieces (Fig. 5-1), of which one piece was thermally modified according to the heat treatment process and the other was used as a reference material.
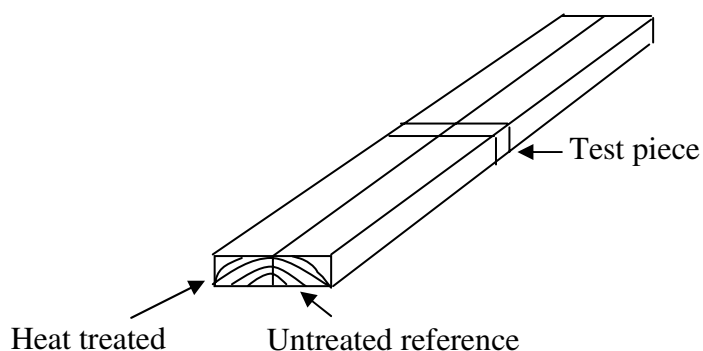
Figure 5-1. Description of the timber used in the heat treatment.

Several Scots pine samples were heat treated in the temperature range of 115-240ºC for different periods. Norway spruce was heat treated at 195ºC; the total duration of the heat treatment was 44 h and the actual time at high temperature was 188 min. Silver birch and European aspen were also treated at 195ºC, but the total duration of the heat treatment was 56 h and the actual time at high temperature was 130 min. European oak was heat treated at 160ºC. The details of the heat treatments are presented in publications I-III and V.

5.1.1 Biodegradation

The tests of the biological resistance of Scots pine against soft and brown rot fungi were performed at VTT according to the standards EN 807 and EN 113, respectively. The details are presented in publication II.

5.1.2 Weathering

Scots pine wood panels were exposed to natural weathering for seven years (1994-2001) in Espoo, Finland. The panels were weathered vertically, heartwood side up, on racks facing south. Weathered thermally modified and unmodified panels (one of each) were selected for spectroscopic analysis. Samples were taken from the surface of the panels. The details are presented in publication III.

5.2 Wood samples for NMR analysis

Heat treated   Untreated reference
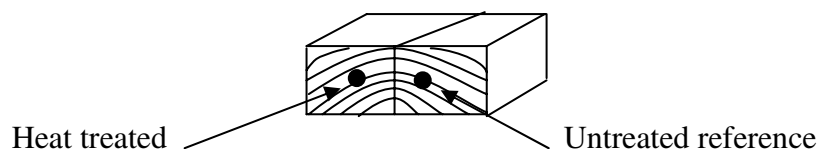
Figure 5-2. Description of the pine wood samples used in the NMR analysis. Cylindrical samples are indicated by the arrows.

Thermally modified and reference wood samples of each species were taken from the same tree trunk. Thermally modified pine wood samples and their reference samples were, in addition, taken from the same annual ring of the wood (Fig. 5-2). Wood samples contained both early wood and late wood. Actual samples were cylindrical cores or small pieces of wood (Fig. 5-2). All samples were moistened with deionised water before being placed into the NMR rotor. This was done because water is known to improve resolution and the signal-to-noise ratio in cellulose spectra.[49,50] The details of the sample preparations are presented in publications I-V.

6 ^{13}C CPMAS NMR SPECTROSCOPIC TECHNIQUES

As described, solid state ^{13}C NMR spectroscopy has been successfully applied in studies of wood and its components. The techniques of proton-carbon cross polarisation (CP), high-power proton decoupling and magic angle spinning (MAS) are combined in solid state CPMAS NMR measurements. In solid state NMR spectroscopy, the advantage is that the samples can be studied in native state without fractionation or isolation of components and all chemical changes in the structure that might occur in chemical treatment are thereby avoided.

6.1 Study of wood

Several NMR studies of the structure of wood and its individual components have recently been reviewed.[51-55] Cellulose, hemicelluloses and lignin, and to some extent extractives, give characteristic signals in the solid state NMR spectrum. Relatively sharp signals are assigned to ordered cellulose or hemicelluloses, while broader background signals are assigned to lignin and disordered hemicelluloses. The ^{13}C CPMAS NMR spectrum of Scots pine is shown in Fig. 6-1. The specific signal assignments for the spectrum of wood are presented in Table 6-1.

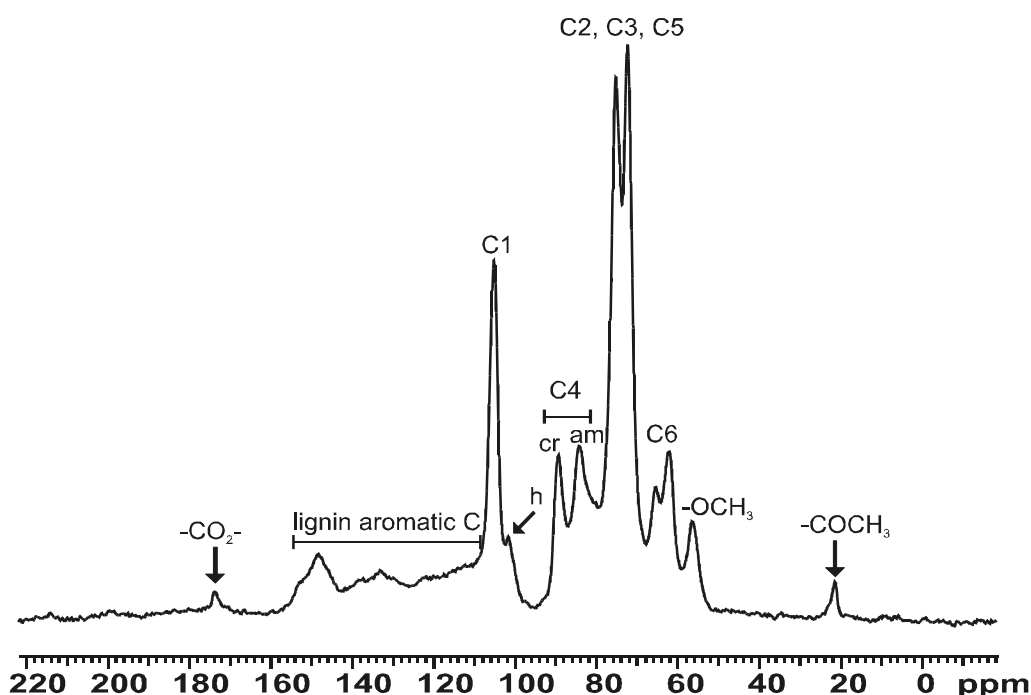***Figure 6-1.*** *^{13}C CPMAS NMR spectrum of Scots pine. Cr refers to crystalline, am to amorphous and h to hemicelluloses.*

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Table 6-1. *Signal assignments for the ^{13}C CPMAS NMR spectrum of wood.*

Chemical shift (ppm)	Assignment
173	CO_2 in acetyl groups of hemicelluloses
153	S 3/5e, G 4e
148	S 3/5f, G 3
146	G 4f
136	S 1/4e, G1e
133	S 1/4f, G1f
120	G 6
116	G 5
112	G 2
105	C-1 of cellulose
102	C-1 of hemicelluloses
89	C-4 of crystalline cellulose
84	C-4 of amorphous cellulose
72-75	C-2/C-3/C-5 of cellulose
65	C-6 of crystalline cellulose
62	C-6 of amorphous cellulose
56	OCH_3 in lignin
21	CH_3 in acetyl groups of hemicelluloses

S = syringyl, G = guaiacyl, e = etherified C-4, f = free phenolic C-4

One drawback of ^{13}C CPMAS NMR measurements of wood is the inability to provide quantitative information about the different components. The spectra contain many overlaps of the various signals of cellulose, hemicelluloses and lignin, due to their similar chemical moieties. Cellulose spectra, however, have been shown to be quantitative due to the chemically equivalent carbons of anhydroglucose units present in cellulose. Cellulose signals in solid state NMR spectra can thus be compared, and quantitative information about cellulose in wood can be obtained, if the signals due to lignin and hemicelluloses are eliminated.

The cross polarisation kinetics differs for the protonated and nonprotonated carbons of wood. The most efficient cross polarisation for CPMAS measurements is obtained by recording the signal intensities of different components in wood as a function of contact time. The contact time is chosen so that intensities of the signals in the spectra develop in a similar fashion. The details of the parameters used in the NMR measurements are presented in publications I-V.

Another problem with solid state NMR analysis is the long measuring times and high costs, which normally limit the number of repeated measurements. At the same time, the relatively large amount of sample needed for NMR measurement (100-300 mg) prevents the problems associated with sample inhomogeneity.

6.1.1 Cellulose

The ^{13}C CPMAS NMR spectrum of wood presented in Fig. 6-1 is dominated by the signals assigned to cellulose. Even though the carbons of the different anhydroglucose units of cellulose are chemically equivalent, they can be separated according, for example, to the different packing of the chains or different conformations. Distinct signals for amorphous and crystalline carbons can be detected. The sharp signals at 105 ppm, 89 ppm and 65 ppm are due to the ordered cellulose C-1, C-4 and C-6 carbons, respectively, while the signals at 84 ppm and 62 ppm are due to the disordered cellulose C-4 and C-6 carbons, as well as to the less ordered cellulose chains of the crystallite surfaces.[52,56,57] The signals at 72-75 ppm are assigned to the C-2,3,5 carbons of cellulose.

6.1.1.1 Determination of cellulose crystallinity

The crystallinity of cellulose, determined as crystallinity index (CrI), was calculated by deconvolution from the area of the crystalline cellulose (86-92 ppm) C-4 signal, a, and the area of the amorphous cellulose (79-86 ppm) C-4 signal, b:[58]

$$CrI = a/(a+b) \cdot 100 \ \% \tag{6.1.}$$

Only the highly ordered cellulose in the interior of the crystallites is considered as crystalline cellulose by this method.[52] The remaining less ordered cellulose, including the fibril surfaces, is referred to here as amorphous. Even though NMR spectral signals in solid samples usually have a Gaussian line shape, Lorentzian line shape was used in most cases because it gave better fit.

The conventional ^{13}C CPMAS spectra of wood contain many signal overlaps. The signals of cellulose are severely overlapped by the signals of hemicelluloses and lignin, and the interfering signals of lignin and hemicelluloses must be removed before CrI can be determined reliably.

6.1.1.2 Application of spin-locking technique

An advanced spectroscopic technique, which provides the spectrum of cellulose without interfering NMR signals of lignin and hemicelluloses, has been described by Newman and Hemmingson[59,60] and Newman.[61] In this technique, differences in proton rotating-frame relaxation times ($T_{1\rho H}$) between the more ordered part (cellulose) and the amorphous matrix (lignin/hemicelluloses) are used to obtain NMR spectra. The subspectra of more ordered cellulose and the amorphous matrix are separated on the basis of the different relaxation

times. The benefit of this purely spectroscopic technique is that changes in the structure of wood components due to chemical treatment of samples are avoided.

The spin-locking pulse sequence presented in Fig. 6-2, referred to as delayed-contact pulse sequence by Newman and Hemmingson,[59] has a spin-lock delay (t_{sl}) between the proton preparation pulse (t_{90}) and the contact time (t_{cntct}), which allows the relaxation of certain of the components. The sequence differs from the standard cross polarisation sequence only by addition of t_{sl}. During the t_{sl}, amorphous lignin and hemicelluloses relax faster than the more ordered cellulose due to the different mobilities of the molecules. The relaxation times of crystalline and amorphous phases are of similar magnitude, however. The selective weakening of the less ordered domains can be enhanced by computing linear combinations of spectra measured with different delays, which isolates the subspectra of cellulosic and noncellulosic domains (Fig. 6-3). Relaxation times of lignin and hemicelluloses are indistinguishable, which means that those components are inseparable.[59]
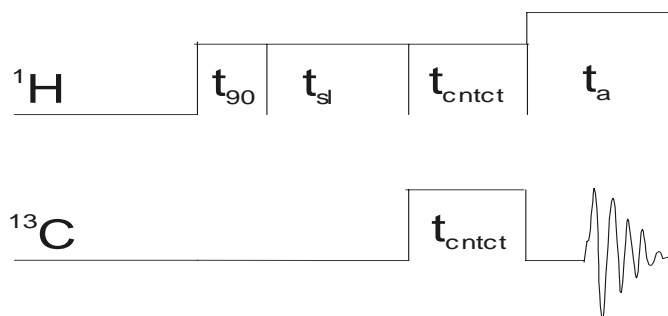

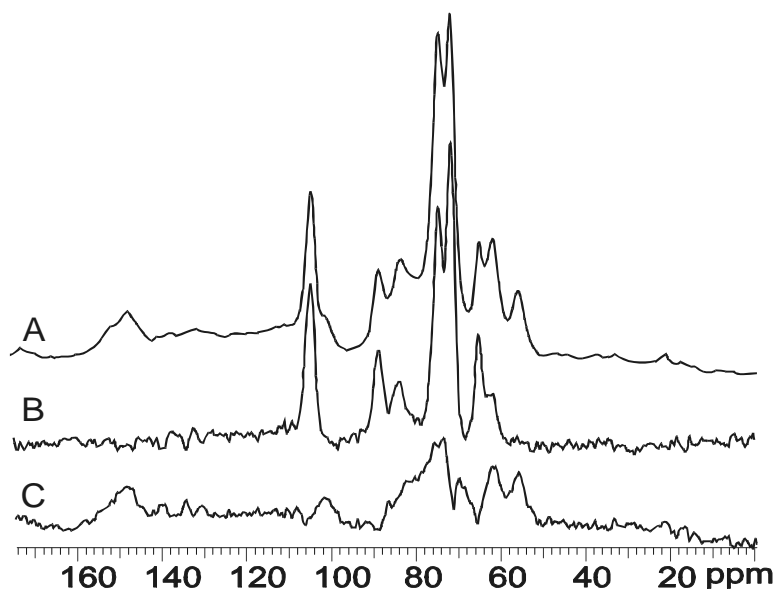*Figure 6-2. Spin-locking pulse sequence.*[59]

Figure 6-3. A) ^{13}C CPMAS NMR spectrum of Scots pine and the separated subspectra of B) cellulose and C) lignin-hemicellulose matrix.

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The procedure for generating a linear combination, described by Newman and Hemmingson,[60] Newman[62,63] as well as Lopes et al.,[64] was applied. The CrI values were calculated from the subspectrum of cellulose (Fig. 6-3B). The spin-locking technique has previously been used to investigate the cellulose crystallinity in wood[39,59,65] and pulp.[66,67]

6.1.2 Hemicelluloses

Investigation of hemicelluloses in the wood matrix by ^{13}C CPMAS NMR is complicated by the strong overlap of the signals assigned to hemicelluloses by the signals of lignin and especially cellulose. Only the signals of methyl (21 ppm) and carboxylic carbons (173 ppm) of acetyl groups attached to hemicelluloses are clear, though weak, in the CPMAS spectrum of wood (Fig. 6-1). In addition to these, the weak shoulder at 102 ppm assigned to hemicelluloses is used in the determination of hemicellulose content.[68,69] This signal is assigned to C-1 of mannose residues in relatively well-ordered glucomannan chains.[39,69,70] Other signals assigned to hemicelluloses have also been reported. The spectrum of more ordered mannan shows a C-4 signal at 81.4 ppm.[69] The signal assigned to xylose in well-ordered xylan chains appears at 83 ppm,[71] but this is severely overlapped by the cellulose C-4 signal in wood spectra making it difficult to interpret.

6.1.3 Lignin

The aromatic carbons of lignin resonate in the ^{13}C CPMAS NMR spectra without nearly any interference from the signals of cellulose and hemicelluloses. The band of signals between 110 and 155 ppm is assigned to different carbons in the lignin structure (Fig. 6-1).[72-74] In addition, the signal at 56 ppm is assigned to methoxyl carbons in lignin. The resolution of the aromatic carbons remains low in conventional ^{13}C CPMAS NMR spectra, however, particularly so in wood samples, and only some general structural features of lignin can be determined. The low resolution can be improved by special techniques that enable more precise study of lignin carbons.

6.1.3.1 Application of dipolar dephasing technique

DD is a useful technique for studying lignin in wood because it suppresses the signals from protonated carbons, so that the quaternary carbons of lignin can be studied in more detail.[75-78] In this technique, the high-power decoupler is turned off for a short while (dipolar dephasing delay, d2) between the contact time (t_{cntct}) and the data acquisition (t_a) (Fig. 6-4). During the d2-delay, the dipolar interactions between protons and carbons cause dephasing of the protonated signals. With a suitable delay time, the protonated carbon signals decay fast and only the quaternary carbons of the lignin aromatic ring appear in the spectrum. The

signals due to fast moving carboxylic carbons of acetyl groups attached to hemicelluloses, as well as the signal due to methoxyl groups, also appear in the DD-spectra.
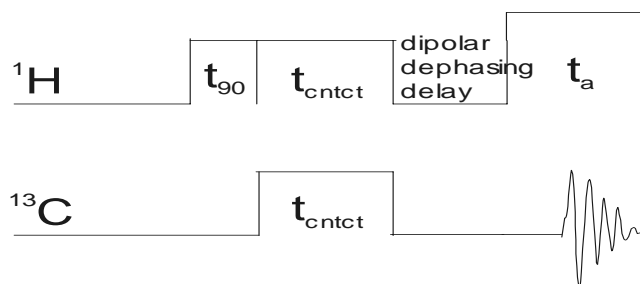
Figure 6-4. Dipolar dephasing pulse sequence.

Examples of the DD spectra of a softwood (spruce) and a hardwood (birch) are shown in Fig. 6-5. The main units in softwoods, G units, give a signal at ~150 ppm assigned to C-3 and C-4 carbons (Fig. 6-5a). The signal at ~133 ppm is assigned to G C-1 and substituted C-5 carbons (Fig. 6-5a). Hardwood lignin contains S as well as G units. S units give signals at 153 ppm and 148 ppm, assigned to different C-3 and C-5 carbons, and a signal at ~135 ppm assigned to C-1 carbons (Fig. 6-5b).
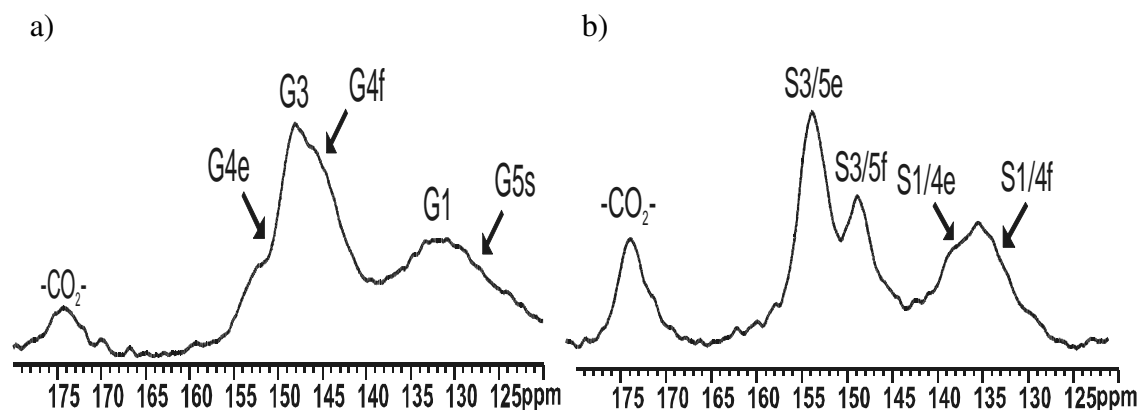
Figure 6-5. Dipolar dephasing spectra of thermally modified a) Scots pine and b) silver birch. G refers to guaiacyl and S to syringyl units; e refers to etherified units, f to free phenolic units and s to substituted structures.

6.1.3.2 Extent of condensation

The extent of condensation of lignin in softwoods and softwood pulps has been estimated by DD technique.[67,79] The signal at ~140-156 ppm in the DD spectrum is assigned to the C-3 and C-4 of G units (A), whereas the signal at ~120-140 ppm is assigned to C-1 and substituted C-5 of G units (B) (Fig. 6-6). The ratio of these signal areas (A/B) provides

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information on the extent of substitution of the aromatic ring and the degree of condensation.[79] The lower the ratio A/B, the higher is the degree of condensation. These same signals are used to determine the ratio of the S and G (i.e. S/G) units in hardwood lignin.[78,80]
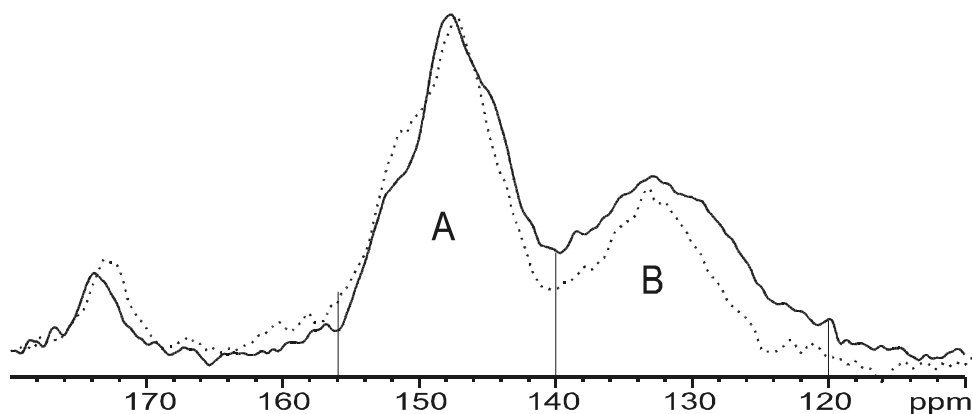
Figure 6-6. *The dipolar dephasing spectra of thermally modified (solid line) and unmodified spruce (dotted line).*

6.2 Principal component analysis in processing NMR spectral data

Principal component analysis (PCA) involves a mathematical procedure that transforms a number of variables into a smaller number of variables called principal components. The first principal component is the combination of variables that explains the greatest amount of variation within samples or sample groups. The second principal component defines the next largest amount of variation and is independent of the first principal component. There can be as many principal components as there are variables.

Multivariate data analysis techniques such as PCA provide a useful way to unravel spectroscopic data.[81] PCA offers a rapid means of identifying and classifying qualitatively great amounts of spectral data in a short time. When spectroscopic data is processed with PCA, the whole spectral range can be taken into account instead of single peak heights or areas, and more accurate characterisation is obtained. Variations within a large amount of spectral data can be characterised by analysing the loading line plots, which in the case of spectral data are the subspectra that show characteristic signals for each sample or sample group. PCA has been applied to the analysis of solid state NMR spectral data previously.[25,82] In this work, PCA was applied to analyse the NMR spectral data of wood samples exposed to biodegradation in order to clarify the behaviour of thermally modified wood in ground contact.

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7 RESULTS AND DISCUSSION

7.1 Chemical structure of wood and thermally modified wood

Thermal degradation of wood involves the deterioration of its individual components: cellulose, hemicelluloses, lignin and extractives. In the following text, the thermal degradation of each wood component is discussed separately, except for extractives, which were not part of the work. The ^{13}C CPMAS NMR spectra of the thermally modified (tm) and reference (ref) softwood samples (Scots pine and Norway spruce) and hardwood samples (European oak, European aspen and silver birch) are shown in Fig. 7-1.

7.1.1 Cellulose crystallinity

7.1.1.1 Effects of thermal modification   *(Publications I & V)*

The deterioration of cellulose in thermal modification is observed as a decrease in the signals at 84 and 62 ppm assigned to disordered cellulose (Fig. 7-1). In every spectrum, the ratio of the relative intensities of the signal at 84 ppm, assigned to the C-4 of disordered cellulose, and the signal at 89 ppm, assigned to the C-4 of highly ordered cellulose, is smaller after thermal modification. The spin-locking technique described in section 6.1.1.2 was applied to determine the effect of thermal modification on the CrI values.[I,V] The results are presented in Table 7-1.

Table 7-1. *Cellulose crystallinity indices (CrI) for wood samples (%). Ref refers to untreated reference sample and tm to thermally modified sample.*[I,V]

Sample	Birch	Aspen	Oak	Spruce	Pine
Ref	41	48	46	54	51
Tm	52	54	53	65	65

The results show that thermal modification enhances cellulose crystallinity. The crystallinity indices are higher for the thermally modified softwood samples of pine and spruce (65%) than for the thermally modified hardwood samples of birch (52%), aspen (54%) and oak (53%). The accuracy of the crystallinity values was estimated to be within ± 3% on the basis of processing of the spectra, which means that the crystallinities of the different thermally modified hardwoods are the same.
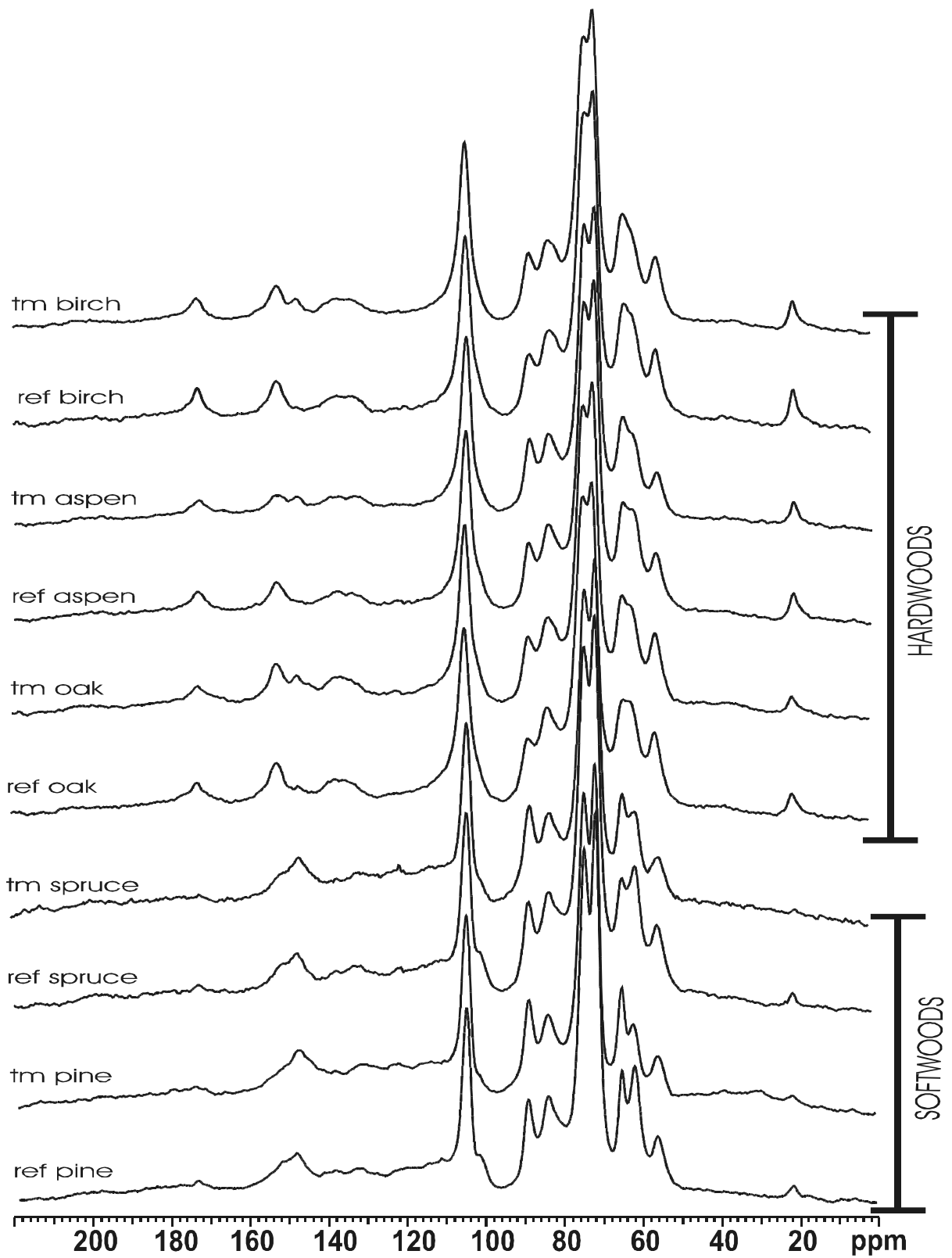
Figure 7-1. ^{13}C *CPMAS NMR spectra of thermally modified (tm) and unmodified reference (ref) samples of birch, aspen, oak, spruce and pine.*

The increase in the cellulose crystallinities with thermal treatment is most likely due to the degradation of the less ordered cellulose chains rather than to any increase in the amount of more ordered cellulose. Additionally, degradation of the less ordered cellulose and hemicelluloses, not the increase in the cellulose crystallinity, is probably the main cause of the reduced strength of thermally modified wood. Arguing in support of this, thermal treatment reportedly has more of a negative effect on the strength of hardwoods than of softwoods,[2,83] even though the degree of cellulose crystallinity is greater for softwoods.

7.1.1.2 Comparison of softwoods and hardwoods   *(Publication V)*

The cellulose crystallinities before thermal modification are higher in softwoods than in hardwoods (Table 7-1).[I,V] CrI for pine is 51% and for spruce 54%, while CrI for birch is 41%, for aspen 48% and for oak 46%. Using spin-locking technique, Newman and Hemmingson,[59] likewise found slightly higher cellulose crystallinities for softwoods (mean value for six softwood samples 57%) than for hardwoods (mean value for five hardwood samples 54%). Newman[63] has further reported the crystallinity and lateral dimensions of crystallites for ten samples using a model in which crystallites have approximately square cross sections. The crystallinity varied between 32 and 49% for samples of plant material obtained without any chemical treatment, between 44 and 47% for unbleached twines and between 65 and 78% for processed and bleached samples. Hult et al.[84] determined the crystallinity of different spruce wood samples by ^{13}C CPMAS NMR. When the relative intensities of the crystalline and paracrystalline cellulose signals were added together the crystallinity values were 30% for compression wood, 28% for earlywood, 33% for latewood, 32% for juvenile wood and 39% for a pulpwood sample. Wood samples have been subjected to a mild chlorite delignification followed by an alkali treatment and a partial removal of hemicelluloses, which is clearly the reason for low crystallinities. Larsson et al.[85] quantified the molecular ordering in birch pulp by non-linear least-squares fitting of the CPMAS spectra. When the results were combined in two categories comprising crystalline and amorphous C-4 signal areas, crystallinity of about 40% was obtained. Using the spin-locking technique, Liitiä[67] found about ten per cent lower cellulose crystallinity values for bleached birch pulp than for bleached pine pulp. The incomplete removal of hemicelluloses contributed to the dissimilarity.

In sum, a wide range of cellulose crystallinity values is reported in the literature. These variations strongly depend on how well lignin and hemicelluloses are eliminated chemically or how successfully the CPMAS spectrum has been spectroscopically divided into the subspectra of cellulose and other wood components.

One relevant factor for the lower cellulose crystallinities of hardwoods than of softwoods in this work is the incomplete removal of xylan by the spin-locking technique. The weak signal of xylan at 83 ppm contributes to the disordered C-4 cellulose signal in the unmodified

hardwood cellulose subspectrum but it is not present in the unmodified softwood cellulose subspectrum. This weak signal of xylan can still be seen at 83 ppm in the hardwood cellulose subspectrum in Fig. 7-2B. The incomplete removal of xylan indicates a similar relaxation behaviour for xylan and cellulose. Possible explanations for this are close association and interactions between these components, or increased relaxation times of xylan due to the ordered structure.[66,67] A slightly weaker signal at 83 ppm is seen in the cellulose subspectrum of thermally modified hardwood in Fig. 7-2D, even though thermal modification degrades hemicelluloses. A similar incomplete removal of the signal of well-ordered glucomannan at 102 ppm by the spin-locking technique has been reported.[39,61,67] The signal of glucomannan could be removed from the spin-locking spectra more completely than that of xylan, however.
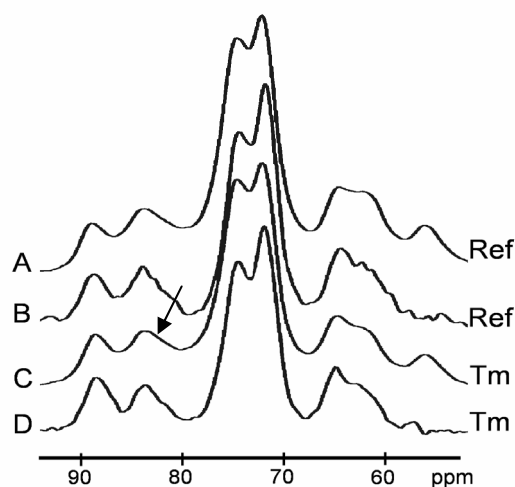
Figure 7-2. ^{13}C *CPMAS NMR spectra of unmodified (A) and thermally modified (C) aspen and cellulose subspectra of unmodified (B) and thermally modified (D) aspen. Ref refers to reference and tm to thermally modified sample. Residual xylan is indicated with an arrow.*

The minor differences in the CrI values of the different softwoods and hardwoods in Table 7-1 derive, in part, from the heterogeneity of wood. The chemical structure varies even within the trunk of a single tree. Variations exist in the structure of cells and pores as well as in the amount and distribution of different cell wall components. The fibre structure and cellulose morphology may differ even within a single fibre. These factors, at least, may affect the degree of cellulose crystallinity. The sample pairs of this work were taken from the trunk of the same tree in order to avoid unnecessary differences in the wood structure.

7.1.1.3 Variations within the stem *(Publication IV)*

The degree of order of cellulose microfibrils was investigated by taking samples of unmodified Scots pine and Norway spruce from different parts of the wood stem: from near the pith and from mature wood, and at different heights of the stem. The cellulose

crystallinities determined by the spin-locking technique for three pine and three spruce samples are presented in Table 7-2. [IV]

Table 7-2. *Cellulose crystallinity indices (CrI) for Scots pine (1 stem) and Norway spruce (2 stems) samples: stem number (Stem), annual ring number from the pith (Year ring) and height.*[IV]

Species	CrI (%)	Stem	Year ring	Height (m)
Pine	54	1	2	8.0
Pine	50	1	7	1.3
Pine	53	1	25	1.3
Spruce	49	2	2	1.3
Spruce	51	2	6	1.3
Spruce	52	3	21	1.3

The CrI values vary between 50 and 54% for pine samples and between 49 and 52% for spruce samples (average 52% for both wood species). As can be seen, the crystallinity of cellulose is similar for the samples taken near the pith and from mature wood and at different heights of the stem. Taking into account the accuracy of the determination ($\pm 3\%$), there is no systematic variation in the crystallinity of cellulose with distance from the pith. Newman[86] has recently reported that, for modelling purposes, it seems reasonable to assume that cellulose crystallinity is homogeneous within a tree and between trees, but the variations in cellulose content cannot be ignored.

7.1.1.4 Comparison of crystallinities determined by ^{13}C CPMAS NMR spectroscopy and X-ray scattering   *(Publication IV)*

The accuracy of the spin-locking technique was evaluated by comparing cellulose crystallinity values obtained by the spin-locking technique and wood crystallinity values obtained by X-ray scattering measurements.[IV] In general, solid state NMR is applied to determine the crystallinity of cellulose and X-ray crystallography to determine the crystallinity of wood. The same pine and spruce samples as presented in Table 7-2, taken from different parts of the wood stem, were investigated by the two techniques. For comparison of the crystallinity values obtained by the two techniques, the crystallinity of the wood was also calculated from the cellulose crystallinity values obtained by NMR using mass fractions 52.2 and 48% for cellulose in *Pinus sylvestris* and *Picea abies*, respectively.[17,87] The wood crystallinity values of 27% for pine and 25% for spruce obtained by NMR are only slightly lower than the values for mature wood obtained by WAXS (31 and 32%, respectively). The absolute error for the crystallinity of wood obtained by WAXS was estimated to be $\pm 4\%$. The agreement between the values obtained by NMR and WAXS is fairly good, therefore, considering that the values used for the mass fractions of cellulose may not be accurate for the early wood samples that were studied.

The mass fraction of cellulose in wood was estimated using the values of cellulose crystallinities obtained by NMR and the values of wood crystallinities obtained by X-ray scattering (see publication IV for details).[IV] Calculated in this way, the mass fraction of cellulose in wood varied from 44 to 58% for pine samples and from 47 to 62% for spruce samples. The accuracy of the values was estimated to be about 10%. Fengel and Wegener[17] and Sjöström[18] give values of 52 and 40% for the mass fraction of cellulose in Scots pine and 46 and 41.7% for Norway spruce. Taking into account the accuracy of the estimations, the values agree well enough with the values in the literature. It can be concluded from this that the spin-locking technique provides a good way to determine cellulose crystallinity.

7.1.2. Hemicelluloses   *(Publications I & V)*

The thermal degradation of wood was shown to start with the hemicelluloses.[I,V] The lower thermal stability of hemicelluloses than of cellulose can be explained by the lack of crystallinity. The relative intensities of the signals of methyl (21 ppm) and carboxylic (173 ppm) carbons of acetyl groups attached to hemicelluloses decrease in every spectrum during thermal modification, indicating the deacetylation of hemicelluloses (Fig. 7-1). The shoulder at 102 ppm on the signal of the cellulose C-1 at 105 ppm, which is assigned to hemicelluloses, is weaker in the spectra of thermally modified softwoods than in the spectra of unmodified softwoods (Fig. 7-1). The shoulder at 102 ppm is not seen at all in the spectra of hardwoods. Softwoods and hardwoods differ in the percentage and composition of hemicelluloses, as noted in section 2.2. Softwoods contain more glucomannans than hardwoods, while hardwoods contain more xylans. The shoulder at 102 ppm can thus be assigned to glucomannans.[68,V] It is reported, however, that hardwood carbohydrates, and especially hemicelluloses (mainly xylan), degrade under milder conditions than softwood carbohydrates.[88] Unfortunately, the signal due to xylan (at 83 ppm) is overlapped by the signal due to cellulose (at 84 ppm) making its interpretation difficult from the conventional ^{13}C CPMAS NMR spectra.

The degradation of hemicelluloses is responsible for a number of modifications of wood properties. The better dimensional stability of thermally modified wood than of unmodified wood is induced by the degradation of hemicelluloses. As a consequence of the degradation of hemicelluloses, the amount of reactive hydroxyl groups in wood is diminished. Hydroxyl groups form hydrogen bonds with water molecules, and the lower content of hydroxyl groups has the effect of decreasing the equilibrium moisture content of thermally modified wood. The reduced strength of thermally modified wood is probably mostly due to the degradation of hemicelluloses, which then no longer solidify the cellulose microfibrils. In continuously moist conditions, the strength of wood is further eroded by the acid (mainly acetic acid) remaining in the wood structure after deacetylation of hemicelluloses,[88,89] which hydrolyses and depolymerises cellulose chains.

7.1.3 Lignin   *(Publications I & V)*

The relative mass fraction of lignin increased during thermal modification in proportion to the relative mass fraction of carbohydrates, being highest for the wood samples treated at highest temperature (Fig. 7-1).[I,V] This change is due to the preferred degradation of carbohydrates relative to lignin.

The DD spectra of thermally modified (tm) and reference (ref) softwood samples (Scots pine and Norway spruce) and hardwood samples (European oak, European aspen and silver birch) are shown in Fig. 7-3.
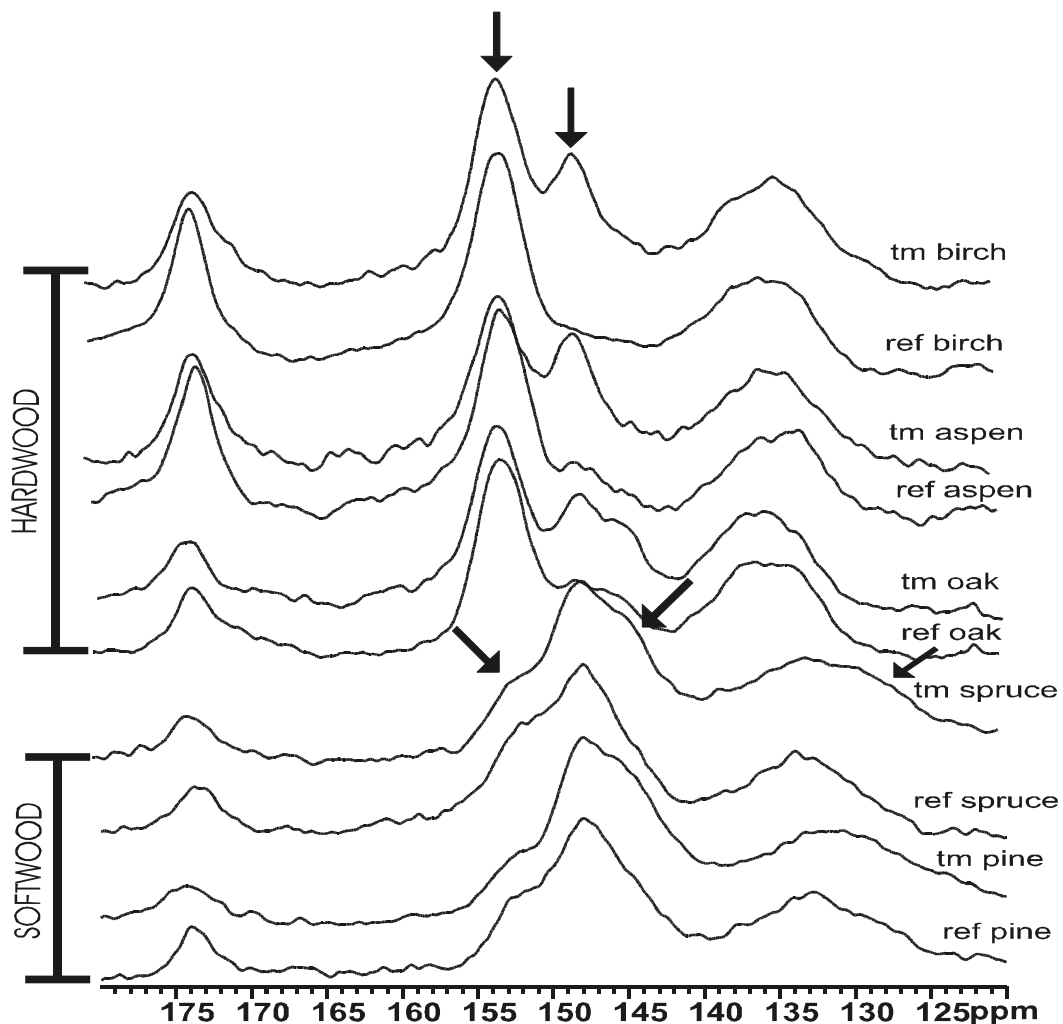

Figure 7-3. *The dipolar dephasing spectra of thermally modified (tm) and unmodified reference (ref) samples of birch, aspen, oak, spruce and pine. Arrows indicate the changes referred to in the text.*

7.1.3.1 Softwoods *(Publications I & V)*

Cleavage of the β-O-4 linkages in lignin is observed in softwoods from shape of the shoulders of the guaiacyl C-3 and C-4 signals at 140-160 ppm, indicated with arrows in Fig. 7-3.[V] The etherified G units in lignin are seen as a shoulder at 153 ppm, while the free phenolic G units in lignin appear as a shoulder at 146 ppm. Lignin contains more free phenolic G units after than before thermal modification, which indicates cleavage of the β-O-4 linkages.

A shoulder at 128 ppm (arrow in Fig. 7-3) appear in the softwood spectra after thermal treatment.[I,V]. This shoulder is assumed to arise from lignin C-5-substituted structures, such as biphenyl (5-5) or diphenylmethane. The appearance of the signal signifies that G units are linked by C-C bonds, and that the content of condensed G structures is increased during thermal modification. Several papers have reported the condensation of lignin at elevated temperatures.[6,23,90,91] The condensation has been interpreted as mainly a diphenylmethane-type condensation.[6,90] The extent of lignin condensation was determined by DD technique, as described in section 6.1.3.2. The A/B ratios were lower in thermally modified softwoods than in unmodified softwoods, indicating higher degree of condensation in the thermally modified softwoods.[I,V] This increase in condensation of lignin is further supported by a slight decrease in the relative intensity of the methoxyl signal at 56 ppm in the ^{13}C CPMAS NMR spectra of softwoods (Fig. 7-1).[I,V,6,19] Demethoxylation of lignin increases crosslink sites within the lignin leading to a more condensed structure. The improved dimensional stability of thermally modified wood may partly be due to crosslinking of lignin, though it is probably mainly due to the degradation of hemicelluloses.

7.1.3.2 Hardwoods *(Publication V)*

Cleavage of the β-O-4 linkages in lignin was observed in the hardwoods after thermal modification.[V] The relative intensities of the signals at 153 and 148 ppm, indicated with the arrows in Fig. 7-3, have been used as a measure of the degree to which β-O-4 linkages in lignin are cleaved in the heat treatment.[22,92] The signal at 153 ppm is assigned to S C-3/5 units that are etherified at C-4, and the signal at 148 ppm to S 3/5 carbons in free phenolic units and to G 3/4 carbons. The majority of the S units in hardwoods appears to contain β-O-4 linkages before treatment, whereas after treatment a substantial part of these linkages are cleaved.[V] This change is seen as an increase of the signal at 148 ppm and a decrease of the signal at 153 ppm in Fig. 7-3. The extensive β-O-4 bond cleavage is probably due to the steam used in the heat treatment process.[21,22,92] Thermal modification and steam partially depolymerise wood lignin by cleaving the aryl ether linkages involving C-4 of S and G units. Clevage leads to the formation of free hydroxyl phenolic groups and α- and β-carbonyl groups. However, only insignificant signs of new carbonyl groups are seen in the NMR spectral region of 165-200 ppm (Fig. 7-1). Probably, at least part of the volatile and water-

soluble products, like depolymerised hemicelluloses and lignin, were leached out of the wood structure with steam during the modification process, and the remaining amount is too small to be detected by solid state NMR.

7.1.3.3 Comparison of softwoods and hardwoods   *(Publication V)*

Softwood lignin is mainly composed of G units, while hardwood lignin is composed of both G and S units. DD spectra of hardwoods and softwoods differ in the signals due to these units.[V] The ratio of the signals after thermal modification reveals that the cleavage of β-O-4 linkages is more extensive in hardwoods than in softwoods. S units are reported to depolymerise to a greater extent than G units under steam treatment.[21,22] However, structural changes in the hardwood lignin are much more evident in the ^{13}C CPMAS NMR spectra than are the changes in the softwood lignin.

Condensation was not observed in hardwoods after thermal modification.[V] Lignin is assumed to condense by the formation of carbon-carbon bonds at C-5 and/or C-3 positions. These bonds cannot be formed between the S units of hardwoods because methoxyl groups occupy most of the C-5 and C-3 positions. The better strength of thermally modified softwoods relative to thermally modified hardwoods[2,83] can partly be explained by the condensation of lignin of thermally modified softwoods.

7.2 Chemical structure of wood and thermally modified wood exposed to outdoor conditions

7.2.1 Biodegradation   *(Publication II)*

Several Scots pine wood samples that had been thermally modified at different temperatures were exposed, along with their reference samples, to soft rot fungus and brown rot fungus (*Poria placenta*).[II] Examples of the weight losses of the samples are shown in Fig. 7-4. In every case, the weight losses of the thermally modified wood samples are smaller than the weight losses of the untreated reference samples. It is clear, therefore, that thermal modification affects the ability of fungi to degrade wood. Weight losses after the attack by fungi are due to decay and chemical changes in cellulose, hemicelluloses and lignin.
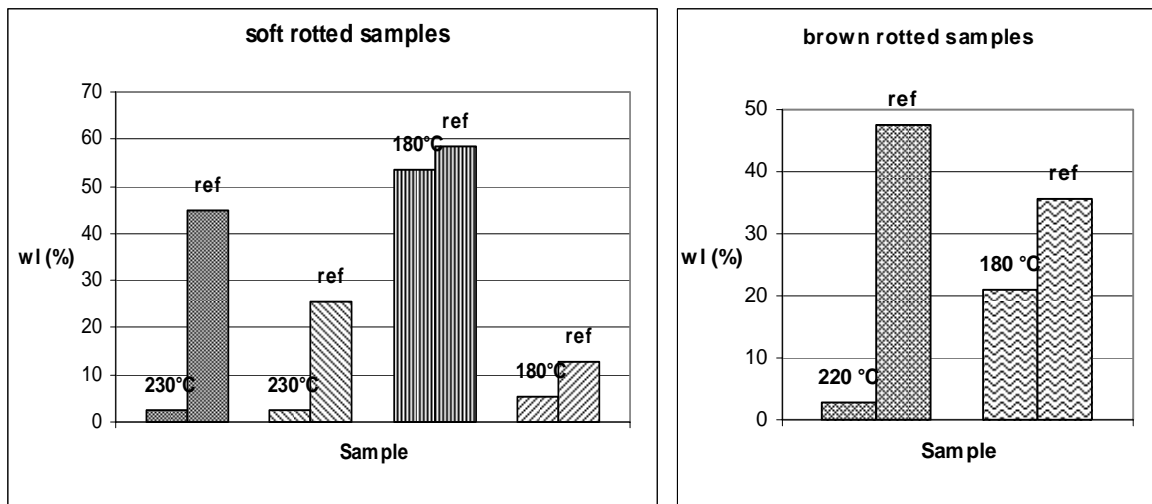
Figure 7-4. *Weight losses (wl) of soft and brown rotted Scots pine samples. 230 ℃, 180 ℃ and 220ºC refer to the temperatures of the heat treatment and ref refers to the corresponding reference sample.*

7.2.1.1 Effects of soft rot fungus

Figure 7-5 shows the ^{13}C CPMAS NMR spectra of a thermally modified pine wood sample (B) and its reference (C) after exposure to soft rot fungus. For comparison, the spectra of thermally modified pine wood sample (A) and its reference (D) not exposed to soft rot fungus are shown in the same figure.

Figure 7-5. *^{13}C CPMAS NMR spectra of the (A) thermally modified, (B) thermally modified and soft rotted, (C) reference and soft rotted and (D) reference pine wood samples.[II]*
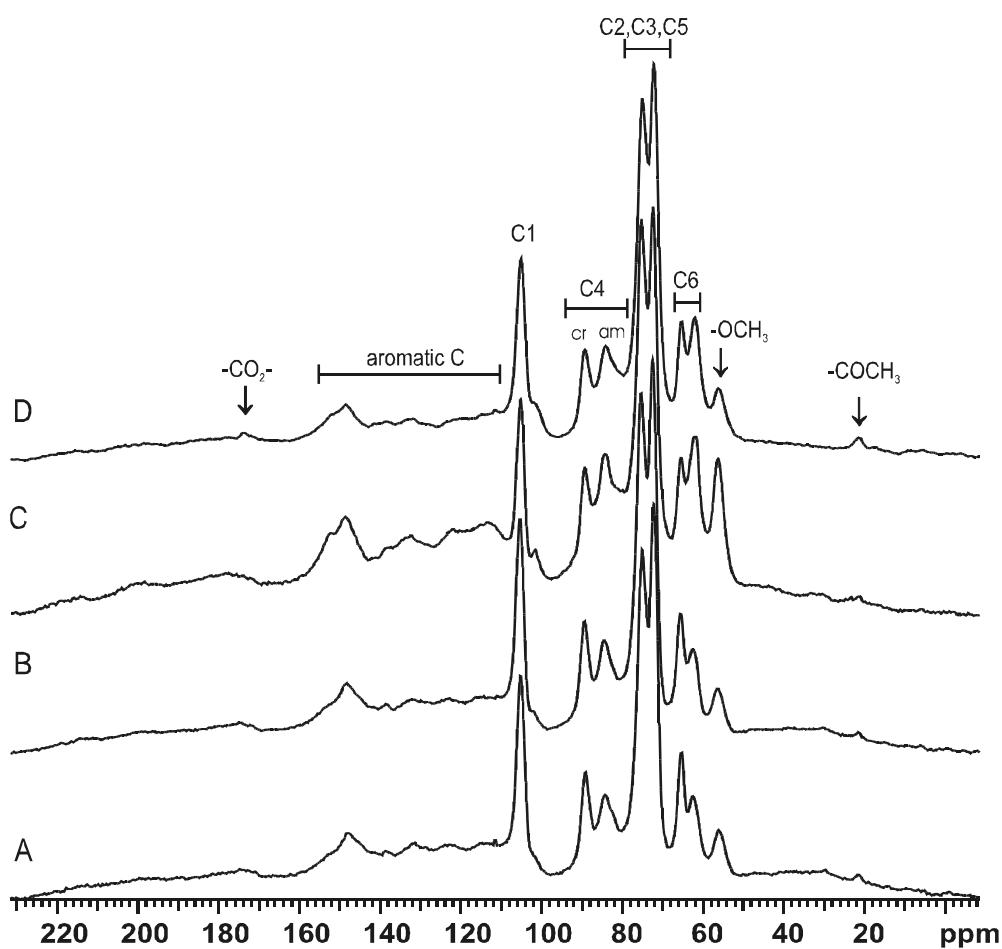
PCA was used to interpret the NMR spectral data of the samples, some of which were exposed to soft rot fungus.[II] The first principal component (p[1]) in the scoreplot in Fig. 7-6a sorts the samples according to their weight losses. The loading line plot of the first principal component in Fig. 7-6b therefore contains information about the rotting pattern of the soft rot fungus: those structures that were enriched in the decay appear, in the loading line plot, as positive peaks, while the structures that were decomposed appear as negative peaks. The second principal component (p[2]) in the scoreplot distinguishes the samples that were thermally modified at 230°C. The loading line plot of the second principal component in Fig. 7-6c therefore contains information about the chemical changes caused by heat treatment at high temperatures. Here the structures that were decomposed in the treatment appear as positive peaks, whereas the enriched structures give negative peaks.
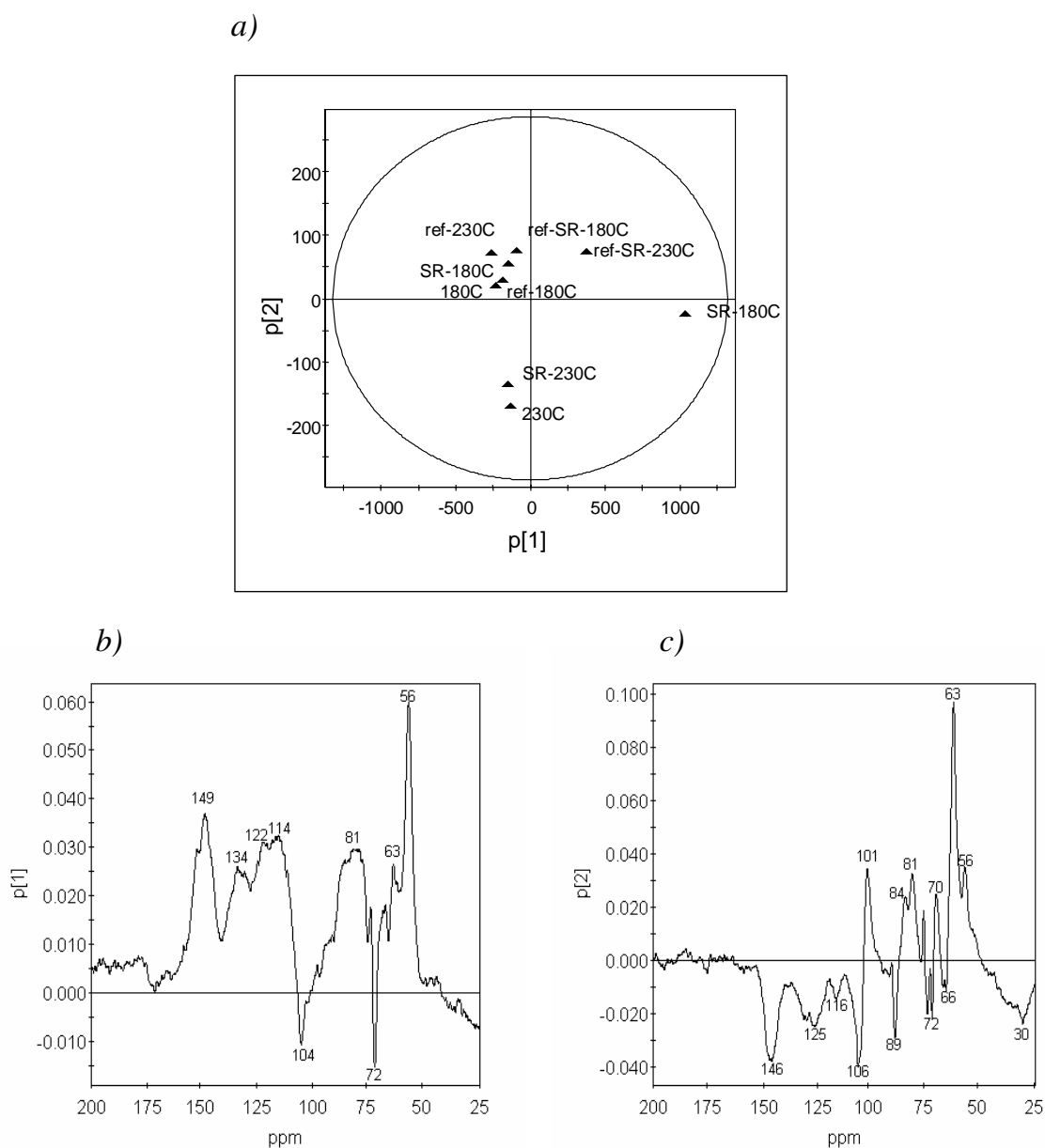
Figure 7-6. a) PCA scatter plot of the first (p[1]) and second (p[2]) principal components of the NMR spectral data of the thermally modified and reference pine wood samples, part of which were exposed to soft rot fungus.[II] Ref refers to reference, SR to soft rotted sample and 180°C and 230°C to the temperatures of the heat treatments. b) Loading line plot of the first principal component (p[1]) and c) loading line plot of the second principal component (p[2]).

A major increase in the relative intensities of the lignin region (110-155 ppm) and the methoxyl signal (56 ppm) relative to the signal intensities of the carbohydrate region is observed in the loading line plot of the first principal component indicating a preferential removal of carbohydrates relative to lignin by the soft rot fungus (Fig 7-6b). The resonance area of the cellulose signals is decreased, which is seen as negative peaks. Soft rot fungus

evidently degrades amorphous and crystalline cellulose to the same extent since the intensities of the signals at 89 and 84 ppm assigned to the crystalline and amorphous cellulose, respectively, do not differ significantly in spectra C and D in Fig. 7-5. The relative intensities of the signals in the carbonyl region (165-200 ppm) are increased (Fig. 7-6b), suggesting the generation of new carbonyl moieties by soft rot fungus.

The loading line plot of the second principal component contains information about the changes in the structure of wood induced by thermal modification (Fig. 7-6c). These changes are discussed in section 7-1.

7.2.1.2 Comparison of the effects of soft and brown rot fungi

The rotting patterns of soft and brown rot fungi (for details, see publication II) appear to be fairly similar.[II] As revealed by PCA analysis, the main difference is that brown rot fungus degrades mainly hemicellulose, while soft rot fungus attacks cellulose more extensively. An increase in lignin content relative to carbohydrates and the generation of new carbonyl structures were observed for both fungi.

The first principal component in the scoreplot distinguished the soft and brown rotted samples according to their weight losses. As presented in Fig. 7-4, the weight losses of thermally modified wood samples are smaller than those of the unmodified reference samples in every case. The results indicate that thermally modified wood is more resistant to biodegradation due to soft and brown rot fungi than is unmodified wood.

7.2.2. Weathering   *(Publication III)*

Thermally modified and unmodified Scots pine wood panels were exposed to natural weathering for seven years.[III] Surfaces of the weathered wood panels became grey, and cracks appeared. The ^{13}C CPMAS NMR spectra of the weathered thermally modified sample (B) and its weathered reference (C) are presented in Fig. 7-7. For comparison, the spectra of the unweathered thermally modified sample (A) and its unweathered reference (D) are shown in the same figure.
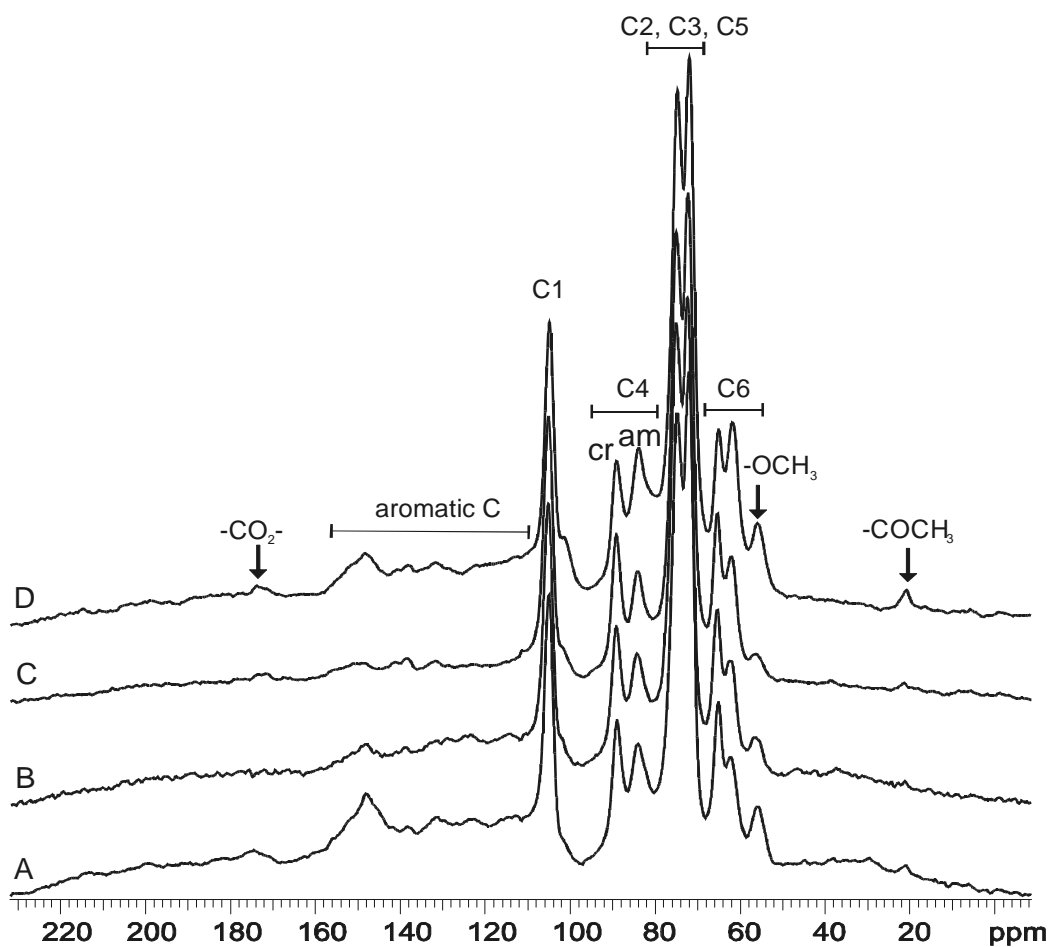
Figure 7-7. ^{13}C *CPMAS NMR spectra of the (A) unweathered thermally modified, (B) weathered thermally modified, (C) weathered reference and (D) unweathered reference pine wood samples.*[III]

The lignin content of the weathered samples decreased dramatically,[III] which is seen as a decrease in the relative intensity of the signals in the lignin region (110-155 ppm) in spectra B, and still more so in C, relative to spectra A and D, respectively. Some structural changes can be seen in the carbohydrate fraction, too. The relative intensity of the signal assigned to hemicelluloses at 102 ppm is diminished in spectrum C relative to spectrum D. This signal is virtually absent from spectra A and B because thermal modification degrades hemicelluloses. The signals of methyl (21 ppm) and carboxylic (173 ppm) carbons of acetyl groups attached to hemicelluloses are also weaker in spectrum B than in spectrum A. These changes indicate degradation of hemicelluloses during weathering. It is assumed that UV light decomposes lignin, and low-molecular-mass lignin fragments that are water-soluble are formed. Water would leach away these lignin fragments, together with hemicelluloses, as water-soluble lignocellulosic materials. The degradation of hemicelluloses suggests that hemicelluloses would be removed along with lignin. Lignin decomposition accelerates the loss of hemicelluloses during exposure to water.

The relative intensities of the signals due to amorphous cellulose at 84 and 62 ppm are also weaker in spectrum C than in spectrum D. However, lignin signals overlap the signals of amorphous cellulose, and the severe degradation of lignin during weathering is responsible for some of the decrease in the signals at cellulose region.

The DD technique is suitable for studying photodegradation by solid state NMR because it suppresses the signals from protonated carbons, revealing the structure of lignin quaternary carbons. The DD spectra of the weathered thermally modified sample and its weathered reference sample are presented in Fig. 7-8. According to the spectra, the lignin content (110-155 ppm) decreases dramatically in weathering, especially in the reference wood sample (Fig. 7-8A). The increase in the condensed lignin structures as a consequence of the heat treatment is seen in spectrum B as a shoulder at 128 ppm. The more crosslinked structure of lignin can partly inhibit UV-light-induced free radical reactions and the formation of low molecular mass degradation products such as quinones.
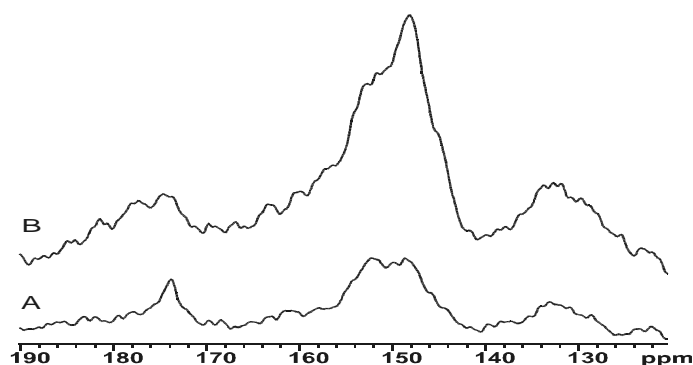

Figure 7-8. *Dipolar dephasing spectra of the (A) weathered reference and (B) weathered thermally modified pine samples.[III]*

The shape and size of the signal at 173 ppm in spectrum B (Fig. 7-8) assigned principally to the carbonyl carbons of hemicelluloses indicates the formation of new carbonyl structures during weathering. No broadening is seen in the DD spectra of the weathered reference sample. New carbonyl structures are still seen in the spectrum of the weathered thermally modified wood sample (Fig. 7-8B), but not in the spectrum of the weathered reference sample (Fig. 7-8A). These results suggest that the weathering products of lignin were leached out with water from the reference sample, whereas in the thermally modified wood they were largely unleachable due to modification of the structures. Evidently, the surface of the weathered thermally modified wood sample was rich in aromatic and conjugated carbonyl structures, whereas the surface of the reference sample was enriched with cellulose. It can be concluded that water, in the presence of UV light, accelerates the degradation of lignin resulting in more destruction of the lignin-hemicellulose matrix in wood cell walls. The results indicate that thermally modified wood is more resistant to natural weathering than is untreated wood. Coating of the surface of heat treated wood is nevertheless recommended.[13,14]

8 CONCLUSIONS

Various solid state ^{13}C CPMAS NMR measurements were performed to investigate the changes that a Finnish industrial-scale heat treatment process brought about in the chemical structure of pine, spruce, birch, aspen and oak. Part of the thermally modified and unmodified wood samples were further exposed to soft and brown rot fungi and weathering.

According to conventional ^{13}C CPMAS NMR spectra, degradation and deacetylation of hemicelluloses occurred in both softwoods and hardwoods during thermal modification. Deacetylation of hemicelluloses leads to liberation of acetic acid, which further catalyses depolymerisation of the less ordered carbohydrates.

Determination of cellulose crystallinity is impossible with conventional ^{13}C CPMAS NMR measurements because the signals of the different wood components overlap. The subspectra of cellulose and the lignin-hemicellulose matrix were separated by the spin-locking technique and linear combination, allowing the cellulose crystallinity to be determined from the subspectrum of cellulose. NMR results showed the CrI values to increase with thermal modification in every wood sample, as a consequence of the preferred degradation of the less ordered carbohydrates. Some differences in the CrI values were apparent for the different wood species. The CrI values were lower for hardwoods than for softwoods both before and after the thermal treatment. Taking the sample pairs from the same tree trunk eliminated unnecessary errors due to the heterogeneity in wood structure. Differences in the CrI values between samples taken from the same trunk were shown to be minor. The accuracy of the spin-locking technique was evaluated by comparing cellulose crystallinity values obtained by the spin-locking technique and the wood crystallinity values obtained by WAXS measurements for the same set of pine and spruce wood samples. The values were in good agreement.

The DD method was applied to study the lignin quaternary carbons and the degree of condensation. Besides the cleavage of the β-O-4 linkages and demethoxylation in both hardwood and softwood lignin, condensation was observed in softwood lignin after thermal modification.

^{13}C CPMAS NMR spectral data were interpreted by PCA to study thermally modified and unmodified Scots pine wood samples exposed to soft and brown rot fungi. Wood samples could be classified according to their weight losses and heat treatment temperatures by PCA. Both fungi caused a drastic decay of the cell wall polysaccharides in unmodified wood samples. Soft rot fungus attacked cellulose more extensively, while brown rot fungus degraded mainly hemicelluloses. The changes in lignin structure were minor. Thermal modification clearly increased the biological resistance of pine.

Conventional ^{13}C CPMAS and DD measurements were applied to examine thermally modified and unmodified Scots pine samples that had been exposed to natural weathering for seven years. The NMR spectra revealed a significant decrease in lignin content of the weathered thermally modified and even more so that of weathered unmodified samples. According to the results, the surface of the thermally modified sample was still rich in aromatic and conjugated carbonyl structures, whereas the surface of the unmodified sample was enriched with cellulose. The structure of the thermally modified wood sample was modified, and the degradation products did not leach out with water as easily as those of the unmodified sample. The weather resistance of thermally modified pine was increased.

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