

Neutron Scattering Studies of Nano-Scale Wood-Water Interactions

By

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To my loving husband and my amazing parents,

Abstract

Understanding and controlling water in wood is critical to both improving forest products moisture durability and developing new sustainable forest products-based technologies. While wood is known to be hygroscopic, there is still a lack of understanding of the nanoscale wood-water interactions necessary for increased moisture-durability and dimensional stability. My PhD thesis focuses on the development and implementation of neutron scattering methods that can provide insight on both the structural and dynamical changes associated with these interactions so that products with improved moisture durability can be developed efficiently.

Using small angle neutron scattering (SANS) and a custom-built in situ relative humidity chamber I studied the anisotropic moisture-induced swelling of wood nanostructure. First, I studied the effects of sample preparation by comparing SANS patterns of wiley milled wood and intact latewood cell walls, and found that scattering from intact wood provides more information about the spatial arrangement of the wood nanostructures inside the cell wall. Comparisons between SANS patterns from earlywood and latewood, show that the higher cell wall density of latewood results in patterns with more pronounced anisotropic features. Then, by measuring latewood loblolly pine sections obtained from the same growth ring and prepared in each of the primary wood planes, I tracked the cellulose elementary fibril spacing as a function of humidity in both intact and partially cut cell walls. These studies showed that even though swelling at the elementary fibril spacing is responsible for the majority of the transverse swelling observed at the S2 level, it is not primary plane dependent. Additionally, there were no differences in the elementary fibril spacing between partially-cut and intact cell walls, except at high humidity where the spacing in partially-cut cells was higher.

SANS was also used to study the effects of two chemical modifications, namely, adhesive infiltration and acetylation, on the wood nanostructure as well as its moisture-induced swelling.

Tangential-longitudinal latewood loblolly pine 0.5 mm thick sections were acetylated or treated with an adhesive (Phenol-formaldehyde (PF) or polymeric methylene diisocyanate (pMDI)) using deuterated or hydrogenated chemicals. Contrast variation experiments on wood modified with deuterated chemicals revealed that PF can infiltrate the regions between the elementary fibrils, while acetylation does not. The moisture-induced swelling of the chemically modified wood was investigated by studying the samples modified with hydrogenated chemicals using SANS and the previously built humidity chamber. These studies revealed that while both PF and pMDI can infiltrate the microfibrils, only PF reduced significantly the swelling at both the elementary fibril and bulk levels. In acetylated samples, the elementary fibril spacing was proportional to the moisture-content of the sample, which was reduced with increasing acetylation. This suggested that the acetylation treatment did not reduce the swelling at the elementary fibril but prevented water from entering the microfibril by modifying the regions surrounding the elementary fibrils.

Using quasi-elastic neutron scattering (QENS) and a custom-built in situ relative humidity sample environment I measured experimentally the (5 – 400 ps) water dynamics inside wood cell walls for the first time and found that there are two types of bound water in the cell wall, namely, slow and fast water. The motion of both water types is well described by a jump-diffusion model, which corresponds to water molecules whose movement follows a stop and go process. Here, the slow water corresponds to water molecules that are highly associated to the wood polymers, whereas the fast water corresponds to water confined inside nanopores within the wood cell wall.

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Nomenclature (in order of appearance)

MC	Moisture content
M_{H_2O}	Mass of water
$M_{OD_{wood}}$	Oven dry mass of wood
Z	Proton number
b	Scattering length
b_a	Absorption scattering length
b_c	Coherent scattering length
b_i	Incoherent scattering length
q	Wave vector transfer
$\vec{k}_f$	Scattered wave vector
$\vec{k}_i$	Incident wave vector
θ	Scattering angle
λ	Wavelength
ΔE	Energy transfer
E_i	Incident neutron energy
E_f	Scattered neutron energy
m_n	Neutron mass

$\hbar$	Reduced Planck's constant
ω	Neutron frequency
ΔSLD	SANS contrast
SLD_x	Scattering length density of constituent x
V_m	Molecular volume
M	Molecular mass
N_A	Avogadro's number
ρ	Density
ϕ_{D_2O}	Volume content of D ₂ O
$\frac{d\sigma}{d\Omega}(q)$	Differential scattering cross section
σ	Microscopic scattering cross section
σ_c	Coherent microscopic scattering cross section
σ_i	Incoherent microscopic scattering cross section
σ_a	Absorption microscopic scattering cross section
$\frac{d\Sigma(q)}{d\Omega}$	Macroscopic scattering cross section
V	Sample volume
$I(q)$	SANS measured intensity
I_{bckg}	SANS background intensity

T	SANS neutron transmission
φ	Neutron flux
ε	Detector efficiency
A	Illuminated area
d_t	Sample thickness
t	Counting time
$\Delta\Omega$	Solid angle
$P(Q)$	SANS form factor
$S(Q)$	SANS structure factor
$\frac{N}{V}$	Number density of particles
V_p	Particle volume
B	Power law prefactor
P	Power law exponent
d	Characteristic length scale
G	Guinier prefactor
R_g	Radius of gyration
s	Dimensionality parameter
$\frac{d^2\sigma}{d\Omega d\omega}$	Double differential scattering cross section

$S(q, \omega)$	Dynamic structure factor
$R(q, \omega)$	Resolution function
$A_0(q)$	Integrated intensity of Delta function
$\delta(\omega)$	Delta function
$A_1(q)$	Integrated intensity of Lorentzian function
$L(\Gamma, \omega)$	Lorentzian function
$B(q)$	QENS background
$S_{el}(q)$	Elastic intensity
$\langle r^2 \rangle$	Average atomic mean square displacement
Γ	Half width half maximum of the Lorentzian function
D	Self-diffusion coefficient
τ	Residence time
EISF	Elastic incoherent structure factor
j_0	Zeroth order Bessel function
R	Radius of the sphere
$QISF$	Quasielastic incoherent structure factor
B_1	Low-q power law prefactor
P_1	Low-q power law exponent
B_2	Mid-q power law prefactor

P_2	Mid-q power law exponent
A_G	Gaussian peak prefactor
q_0	Gaussian peak position
σ	Gaussian peak width
B_3	Constant background
ASE	Anti-shrink swelling efficiency
S_{mod}	Volumetric swelling coefficient of treated wood
S_0	Volumetric swelling coefficient of untreated wood
S	Volumetric swelling coefficient
$V(RH \text{ or } MC)$	Wood swollen volume at a specific RH or MC
V_{OD}	Oven dry volume
A_D	Area of Delta function
$L_1(q)$	Area of narrow Lorentzian
Γ_1	Half-width half maximum of narrow Lorentzian
$L_2(q)$	Area of broad Lorentzian
Γ_2	Half-width half maximum of broad Lorentzian
$C(q, \omega)$	Linear background
D_1	Self-diffusion coefficient of the narrow Lorentzian
τ_1	Residence time of the narrow Lorentzian

D_2	Self-diffusion coefficient of the broad Lorentzian
τ_2	Residence time of the broad Lorentzian
c_1	Immobile fraction
a	Radius of confinement obtained from EISF
FQISF_1	Fractional QISF of the narrow Lorentzian
FQISF_2	Fractional QISF of the broad Lorentzian

List of Abbreviations (in alphabetical order)

1D	One dimensional
2D	Two-dimensional
ASE	Anti-shrink efficiency
BASIS	Backscattering spectrometer
Bio-SANS	Biological-small angle neutron scattering
CMP	Contrast matching point
D2O	Heavy water or deuterated water
DOE	Department of Energy
dPF	Deuterated phenol-formaldehyde
EF	Elementary fibril
EISF	Elastic incoherent structure factor
EQ-SANS	Extended q-range - small angle neutron scattering
FPL	Forest products laboratory
FSP	Fiber saturation point
GDP	Gross domestic product
HFIR	High flux isotope reactor
IPN	Interpenetrating polymer network
LPSD	Linear position sensitive detector

MC	Moisture content
MD	Molecular dynamics
MFA	Microfibril angle
MSD	Mean square displacement
MW	Molecular weight
ORNL	Oak ridge national laboratory
PF	Phenol-formaldehyde
PI	Phenyl isocyanate
pMDI	Polymeric methylene diphenyl diisocyanate
QENS	Quasielastic neutron scattering
QISF	Quasielastic incoherent structure factor
RH	Relative humidity
RL	Radial-longitudinal
SANS	Small angle neutron scattering
SAXS	Small angle x-ray scattering
SDD	Sample to detector distance
SLD	Scattering length density
SNS	Spallation neutron source
SSD	Sample to source distance

T	Transverse
TL	Tangential-longitudinal
TOF	Time of flight
WPG	Weight percentage gain

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Chapter 1. Introduction

Researchers seek sustainable, “green,” wood-based (lignocellulosic) materials as viable alternatives to non-sustainable materials including petroleum-based polymers and old growth wood. Sustainable lignocellulosic materials can mitigate carbon footprints, reduce environmental pollution,^{1–3} and support healthy economies in rural areas where trees are grown, harvested, and processed. Our ability to develop and expand new markets for lignocellulosic materials depends on how well we can control their properties and tailor them to our needs. Research initiatives to develop sustainable technologies using lignocellulosic material include green electronics from cellulose nanofibers films⁴, cellulose nanocrystal substrates for solar panels⁵, bio-fuels,⁶ and industrial chemicals⁷.

The forest products industry currently accounts for about 5% of the annual gross domestic product (GDP) in the US.⁸ Markets for lignocellulosic materials are expanding. However, one of the key limitations for wood-based materials is the interaction of wood with water: moisture durability is a major concern. While it is known that high levels of moisture in wood can lead to shortened service life due to early degradation, including failure of wood-adhesive bond lines, the nanoscale mechanisms of water uptake and moisture durability are not sufficiently well understood. There are numerous commercially available treatments that improve the moisture-durability and/or dimensional stability of forest products, but these are often developed empirically. How they work is not well understood. Researchers interested in developing new methods are hindered by the lack of experimental data on the moisture-induced swelling of wood at the nanoscale before and after it is modified.

A connection between absorption of water molecules at the molecular scale and changes in properties at bulk wood level is still needed. Furthermore, tools that can provide insight on the state of bound water in the cell wall are also needed to validate the models proposed on how

moisture-sorption occurs at the molecular scale based on Molecular dynamics (MD) simulations in recent years.^{9–11}

1.1 Structure and chemical composition of wood

Wood is a hierarchical material with different levels of structure ranging from polymers to cells to growth rings. At the macro scale, wood is an orthotropic material with three primary planes: tangential-longitudinal (TL), radial-longitudinal (RL), and transverse (T), all shown in Figure 1.1a. Growth rings are easily seen, and each growth ring consists of earlywood and latewood bands. Compared to earlywood, latewood generally possesses a higher number density of cells and the cells have thicker walls. In softwood such as pine the most common cell type is the tracheid, which accounts for over 90% of the softwood volume¹². Ray cells account for most of the remaining volume and are oriented perpendicular to the growth rings in the transverse plane to provide radial transport (Figure 1.1b). Tracheids, which are held together by the lignin-rich middle lamella, are several millimeters long in the longitudinal direction and have a hollow center called the lumen. The tracheid cell wall is typically about 5 to 10 μm thick¹³ and is made of a primary and three secondary cell wall layers (S1, S2, and S3), Figure 1.1c. The relative contribution of each cell wall layer to the total thickness is listed in Table 1.1.

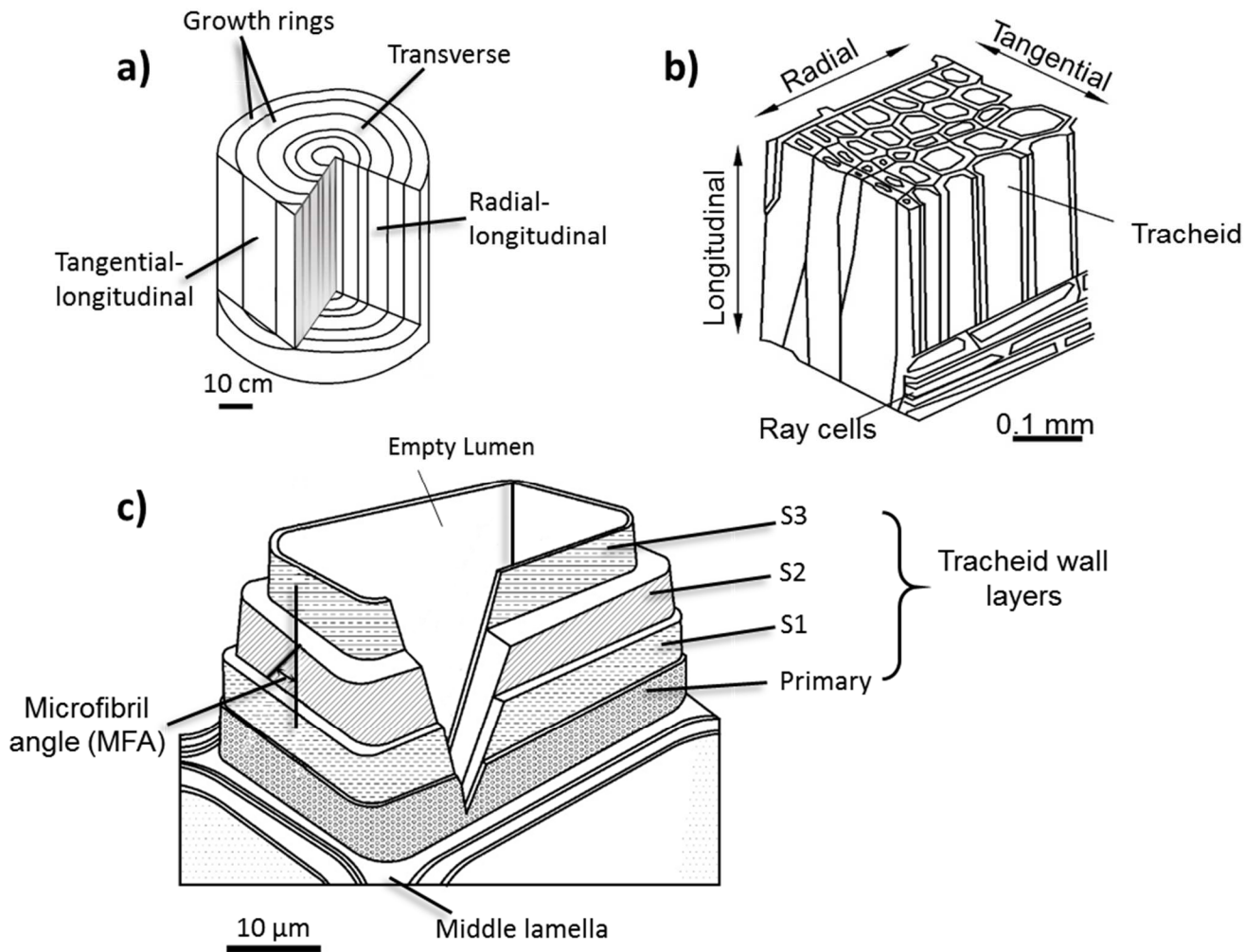


Figure 1.1 Breakdown of wood structure from the a) bulk to the b) cellular to the c) cell wall.

Table 1.1 Relative contribution of each cell wall layer to the total cell wall thickness. Percentages correspond to tracheids in southern yellow pine latewood. ¹⁴

ESTIMATED RELATIVE CELL WALL THICKNESS%	
S1	20%
S2	66%
S3	14%

Secondary cell walls consist of parallel semicrystalline cellulose microfibrils helically embedded in a matrix of lignin and hemicelluloses. The approximate chemical composition of each cell wall layer is listed in Table 1.2. The orientation of the cellulose microfibrils in each layer is quantified

using the microfibril angle (MFA), which is the angle at which the cellulose microfibrils are tilted with respect to the longitudinal tracheid axis. In the S1 and S3 layers, the MFA varies substantially within a given layer¹², and the values reported are high, typically between 50 and 70° for the S1 and over 70° for the S3. Whereas the MFA in the S2, which is the thickest and most dominant layer, is typically between 5 and 30° and shows little variation within a growth ring.

Table 1.2 Chemical composition of each secondary cell wall layer.¹⁴ Percentages correspond to tracheids in southern yellow pine latewood.

Secondary cell wall layer	<i>Cellulose</i>	<i>Hemicelluloses</i>	<i>Lignin</i>
S1	30.0%	18.3%	51.7%
S2	54.3%	30.6%	15.1%
S3	13.0%	87.0%	~0%

Different models have been proposed for the arrangement of the cellulose microfibrils in the S2 layer, Figure 1.2a, including random organization in the transverse plane^{15,16} and alignment parallel to the cell wall thickness¹⁷. Although the detailed structure of the S2 is still debated, most evidence suggests that in the transverse plane the microfibrils are organized in concentric laminae^{18,19}. Several models have also been proposed to describe the organization of the microfibrils in the longitudinal plane, Figure 1.2b, including: parallel to the tracheid axis^{20–22} and a trellis structure²³ but it is not clear yet which is the most likely arrangement.

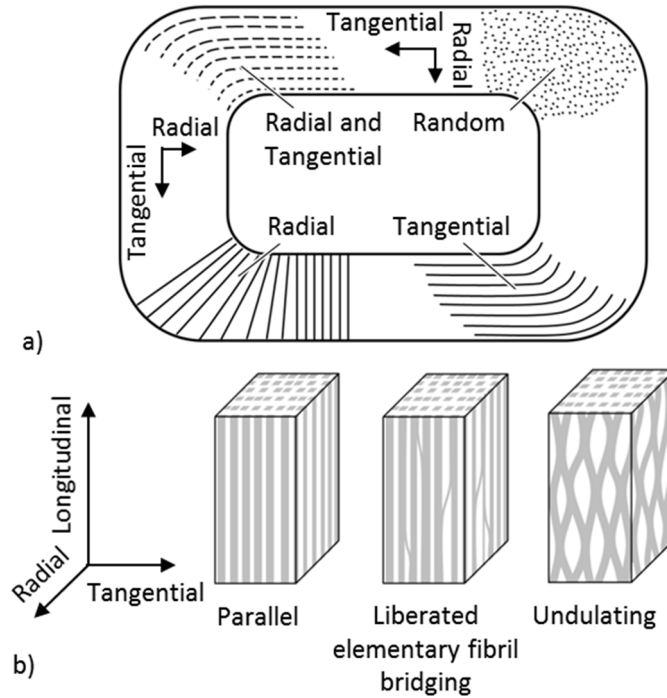


Figure 1.2 Proposed orientations of cellulose microfibrils in S2 layer in the a) transverse and b) longitudinal planes.

While the size of the cellulose microfibrils is not known, most diffraction studies have suggested that their size lies between 16 and 20 nm.^{18,24,25} Microfibrils are composed of elementary fibrils, that are approximately 3 nm across²⁶, and some models have suggested the existence of less ordered regions between them²⁷, Figure 1.3. These less ordered regions are thought to consist of amorphous cellulose and hemicelluloses^{28,29}. Both microfibrils and elementary fibrils have a longitudinal twist^{27,30,31}, which is shown in Figure 1.3. The total number of cellulose chains in the elementary fibril cross section is still debated. For decades it was thought that the total number of chains in the elementary fibril was 36; however, recent scattering experiments suggest that the total number of chains is most likely between 18 to 24^{25,32–34}.

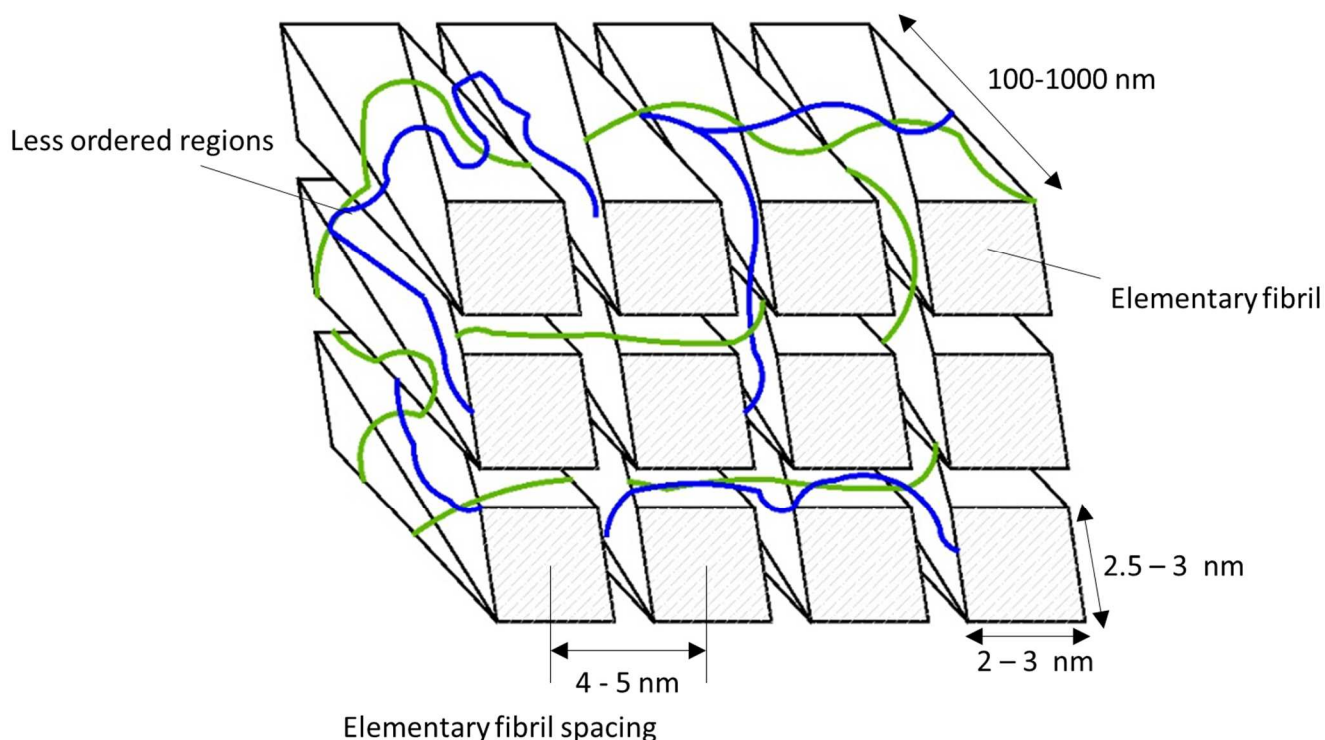


Figure 1.3 Simplified schematic view of the structure of the cellulose microfibril showing a 3 x 4 array of cellulose elementary fibrils with some less ordered regions between the elementary fibrils.²⁸ Schematic was adapted from Martinez et al.³⁵

1.1.1 Wood polymers chemistry and accessibility

Wood cells walls are made of three primary polymer components: cellulose, which can be crystalline or amorphous, hemicelluloses and lignin. A brief background on the chemistry each one of the wood polymers is discussed in detail below.

Cellulose, one of the most abundant polymers on Earth, is the linear polysaccharide shown in Figure 1.4, whose primary unit is the cellobiose. Each cellobiose unit is comprised of two glucose units linked by a $\beta(1 \rightarrow 4)$ glycosidic bond.³⁶ Cellulose, whose chemical formula is $(C_6H_{10}O_5)_n$, has a high degree of polymerization that ranges between 8000 and 10000.

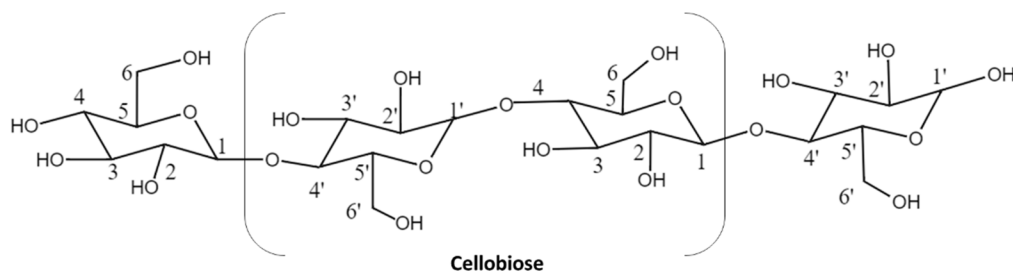


Figure 1.4 Molecular structure of cellulose showing the cellobiose unit, indicated by brackets³⁷. Drawn with ChemDraw Direct.³⁸

Cellulose has been found in at least 6 different crystalline forms, but only two are found naturally in higher order plants, namely I_α and I_β .³⁹ The main difference between these two crystal allomorphs is the hydrogen bonding and the stacking of the crystals. In the triclinic I_α , there is only one cellulose chain per crystal, and the crystals are stacked up due to glucose units that vary slightly in conformation. Each monoclinic cellulose I_β crystal is made of two alternating cellulose chains held together by intramolecular hydrogen bonds; and its crystals are stacked together in parallel held by inter-molecular hydrogen bonds. Crystallographic projections of cellulose I_α and I_β are shown in Figure 1.5. Spectroscopy studies have suggested that both forms of cellulose I can coexist in the native plants, which leads to stacking faults that increase the broadening of secondary diffraction lines.^{40,41} However, the main equatorial peaks are not affected by these stacking faults and most diffraction from wood can be well described by a single crystalline phase of cellulose I_β .

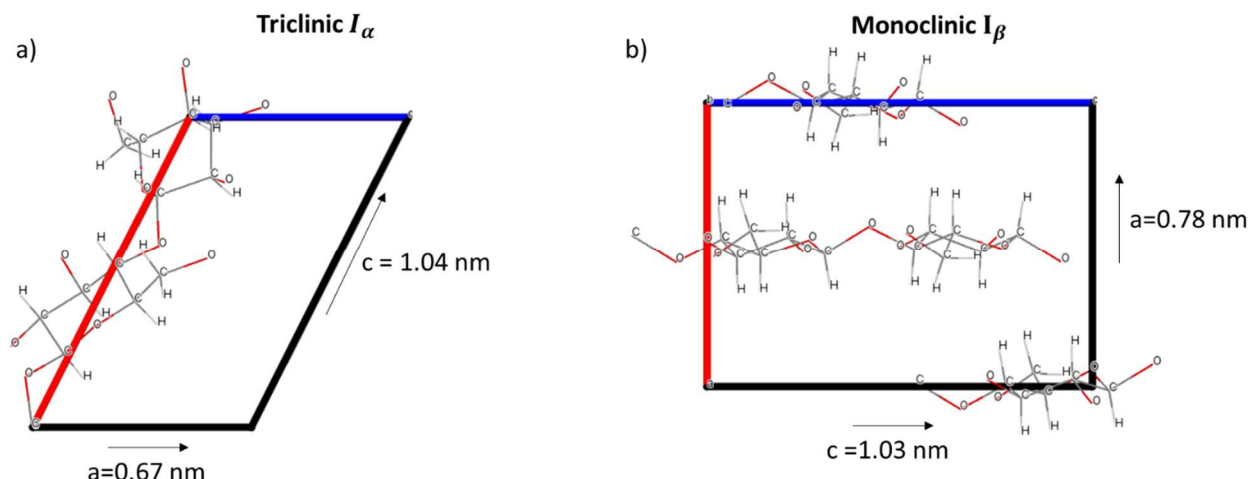


Figure 1.5 Projections along the *ac* plane of the crystalline structures of cellulose: a) triclinic I_α and b) monoclinic I_β . Redrawn using Mercury from ^{40,42}

Cellulose monoclinic crystals have lattice constants of $0.79 \times 0.835 \times 1.03 \text{ nm}$.^{40,43,44} The degree of crystallinity can vary across wood species and even within a wood cell wall but typically about 40% of the cellulose in pine is crystalline. The remaining cellulose is considered amorphous or para-crystalline, which is an intermediate phase between I_α and I_β , and is usually characterized by higher lateral disorder and mobility than crystalline I_α and I_β .^{25,45}

Hemicelluloses are a group of polysaccharides with a lower degree of polymerization than cellulose. These polysaccharides mainly consist of pentose and hexose primary units whose general formulas are $\text{C}_5\text{H}_8\text{O}_4$ and $\text{C}_6\text{H}_{10}\text{O}_5$, respectively. More specifically, the main primary units found in the hemicelluloses are D-xylose, L-arabinose, D-glucose, D-galactose, D-mannose, D-glucuronic acid, 4-methyl-D-glucuronic acid and D-galacturonic acid, which are shown in Figure 1.6. Hemicelluloses are made of different combination of these primary units, and the exact composition varies across the cell wall.⁴⁶ For instance, in softwoods xyloglucan is found in the primary wall, while in the secondary cell wall it is absent or only a minor component.⁴⁷

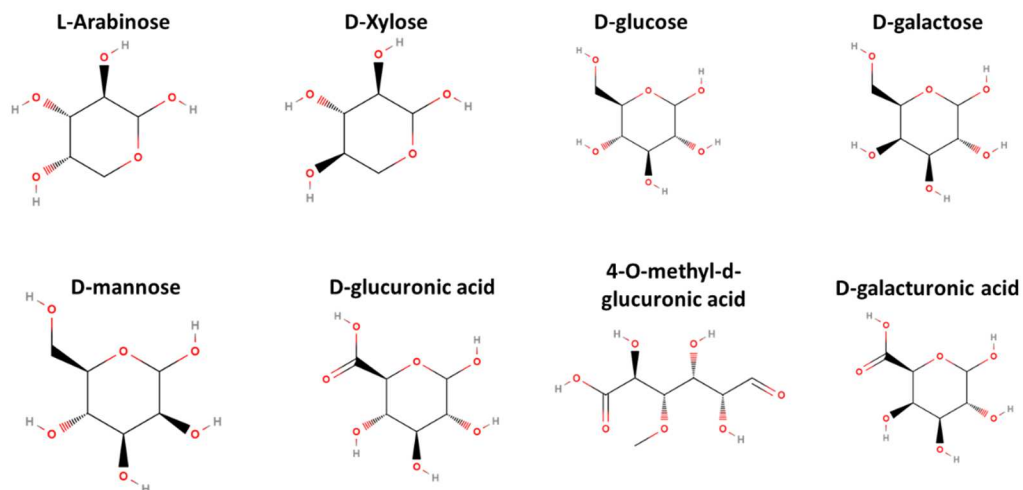
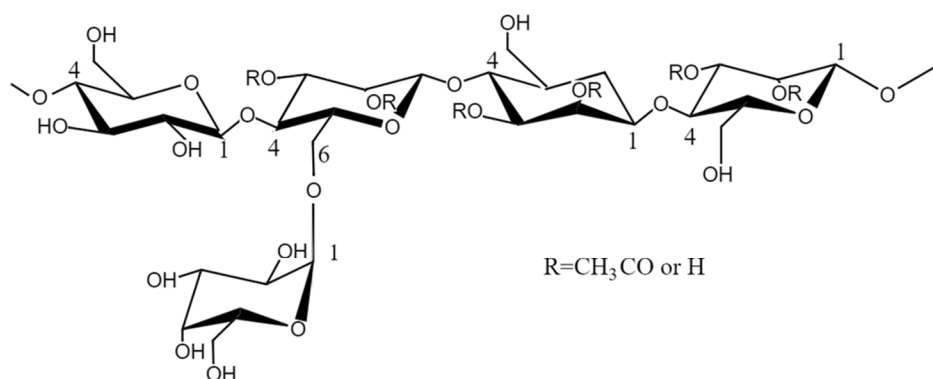


Figure 1.6 Primary monomer units of hemicelluloses. Drawn using Molview⁴⁸.

The major hemicellulosic components found in the secondary cell wall, namely, partially acetylated galactoglucomannans and arabinoglucuronoxylan are shown in Figure 1.7.¹⁴

a) Glucomannan



b) Xylan

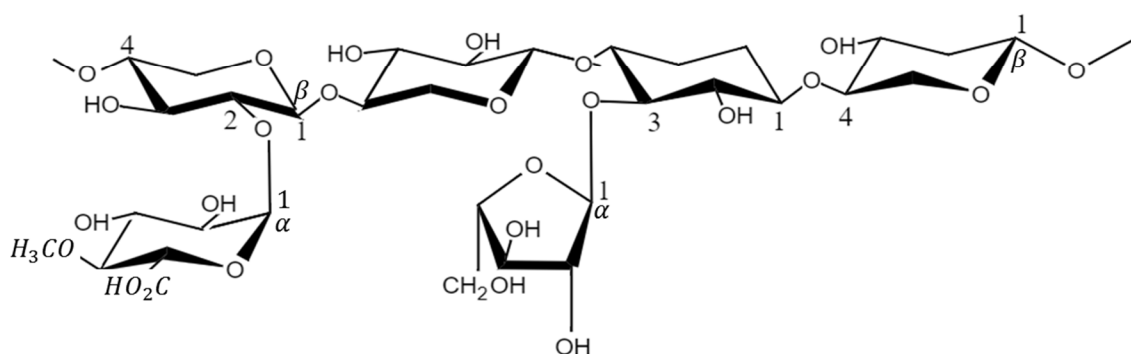


Figure 1.7 Main hemicelluloses found in softwood: (a) Glucomannan and (b) Xylan. Illustration was redrawn from ^{14,49}using ChemDraw Direct.³⁸

From a solubility standpoint, the hemicelluloses act as a bridge between the lignin, discussed later, and the cellulose.⁵⁰ Different hemicelluloses have different levels of affinity with cellulose and lignin. For instance, xylans can interact with cellulose⁵¹ but associate mostly with lignin⁵²; whereas glucomannans have a higher affinity to cellulose than xylans.⁵²

Lignin is a three-dimensional aromatic polymer network. It is composed of phenylpropane units, including coniferyl alcohol, sinapyl alcohol and p-coumaryl alcohol, that are linked by ether and carbon-carbon bonds.^{36,53} These basic units as well as a schematic of the softwood lignin structure is shown in Figure 1.8.

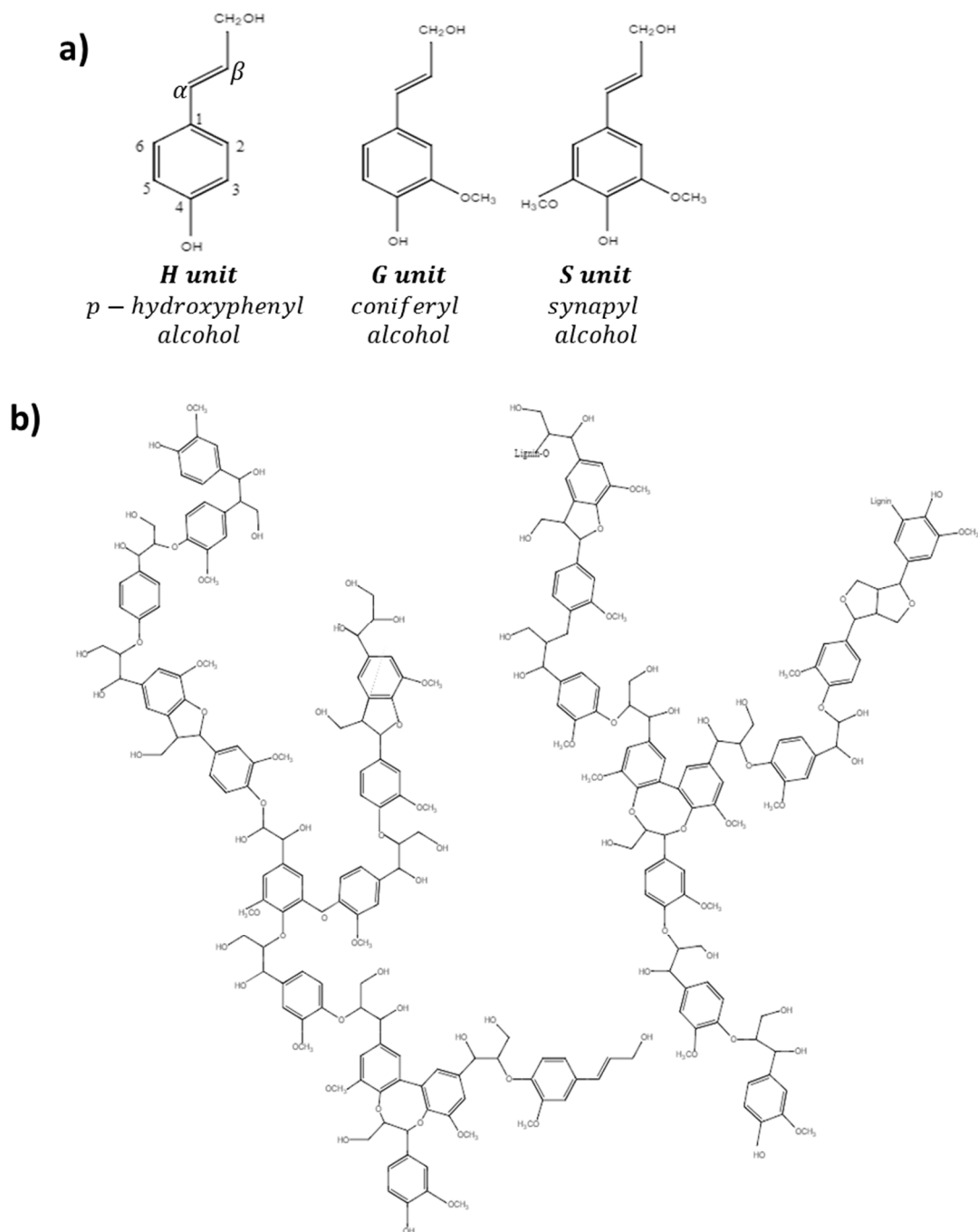


Figure 1.8 Lignin structure. (a) Monolignols, primary precursors of lignin. Redrawn using ChemDraw from⁵³. (b) Schematic of lignin in softwood. Redrawn from⁵⁴ using ChemDraw³⁸.

Recent evidence suggests that the supramolecular structure of lignin is determined by the space available when lignin is deposited during cell wall formation and growth, hence, middle lamella

lignin differs from secondary cell wall lignin.⁵⁵ In the middle lamella, lignin forms 3D globules made of folded chain oligolignols. In the secondary cell wall, lignin can be found in tubular structures surrounding the hemicellulose-coated microfibrils. MD simulations suggest that the aromatic rings of lignin align themselves parallel to the cellulose fibrils, although this order seems limited to the first monolayer of lignin.⁵⁶

1.2 Wood and water relations

The presence of accessible hydroxyl groups and other polar chemical groups in the wood polymers makes wood hygroscopic.⁵⁷ FTIR has been used to study how accessible wood is to D₂O vapor, and it has been found that only about 43% of all hydroxyl groups in earlywood are accessible to D₂O vapor. Because of its high degree of crystallinity, most wood cellulose is inaccessible to water. Only the surface chains of cellulose elementary fibrils are considered accessible to water, which amounts to only 33% of the total hydroxyl groups in cellulose or 6.1 mmol/g.⁵⁷ Hemicelluloses are much more accessible than cellulose. In the hemicelluloses, it is assumed that nearly all of its hydroxyl groups are accessible because of its lack of crystallinity,⁵⁸ and the estimated concentration of available hydroxyl groups ranges from 8.6 to 18.8 mmol/g in glucomannan, whereas in xylan, it is 14.4 mmol/g.⁵⁷ Since lignin is also a non-crystalline polymer it is also expected that most of its hydroxyl groups will be accessible,⁵⁸ and the concentration of accessible hydroxyl groups in is about 7.5 mmol/g⁵⁷, which is comparable to the theoretical accessibility of cellulose. A summary of the accessibilities of all wood polymers is shown in Table 1.3.

Table 1.3 Summary of accessibility of each wood polymer⁵⁹

	Accessible Hydroxyl groups %	
	mmol/g	%
<i>Cellulose</i>	6.1	33
<i>Glucomannan</i>	8.6 - 18.8	~100
<i>Xylan</i>	14.4	~100
<i>Lignin</i>	7.5	~100

In wood science, the amount of water in wood is quantified using the moisture content (MC), which is defined as

$$MC\% = \frac{M_{H_2O}}{M_{OD_{wood}}} * 100 \quad \text{eq. 1.1}$$

where M_{H_2O} is the mass of water and $M_{OD_{wood}}$ is the oven dry mass of wood. The MC of wood is known to depend on temperature and relative humidity (RH). Hence, the RH dependence of the MC is quantified by measuring the sorption isotherm, Figure 1.9, where the mass of added water is monitored as the RH is increased or decreased while keeping a constant temperature. The MC is known to be hysteretic as a function of RH, and measurements taken on the desorption side of the isotherm are typically higher.

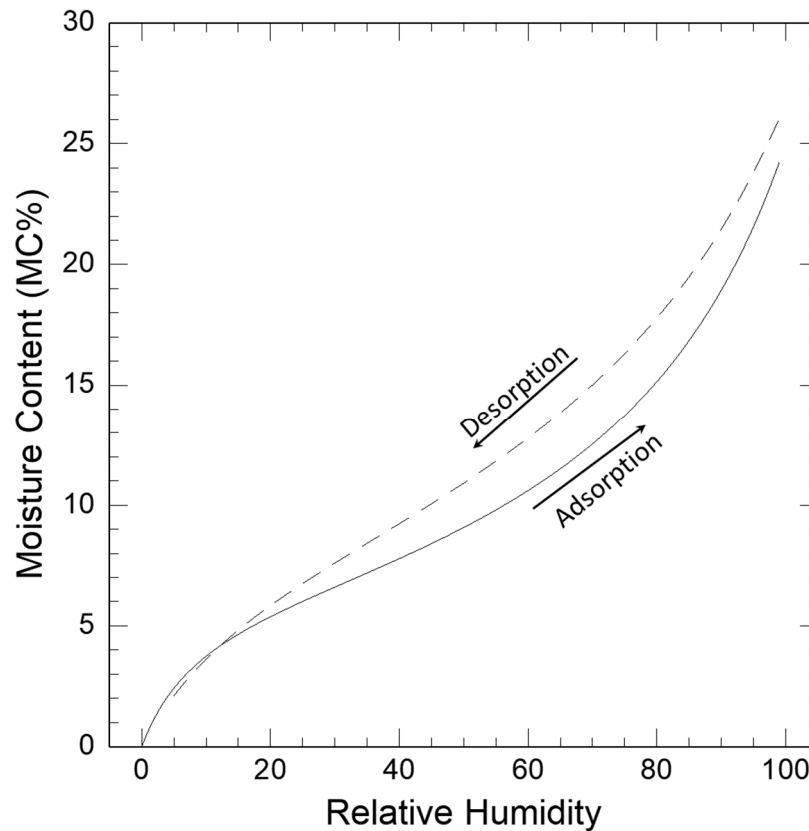


Figure 1.9 Typical adsorption and desorption isotherms of loblolly pine held at 25°C.

At low MC's water exists only as bound water, which is held within the wood polymers by intermolecular forces. As MC increases, free water can begin to form inside the wood cavities, such as pits and lumina, however, this only occurs above the fiber saturation point (FSP). Based on how it is defined and measured, the FSP lies between 30 and 40% MC.^{57,60-62} While the definition and implications of the FSP is an active area of research, the FSP is generally defined as the point where free water starts to form in wood.

Water is expected to exist in all wood polymers except for crystalline cellulose. However, the exact amount of water absorbed by the polymers when they are in the cell wall is not known. While the moisture content of the in-situ wood polymers is not known, it is often assumed that their sorption behavior would be like the extracted materials. Sorption isotherms from the extracted wood

polymers that are hygroscopic are shown in Figure 1.10. Lignin is the least hygroscopic polymer in the cell wall, while, the hygroscopicity of the polysaccharides is comparable.

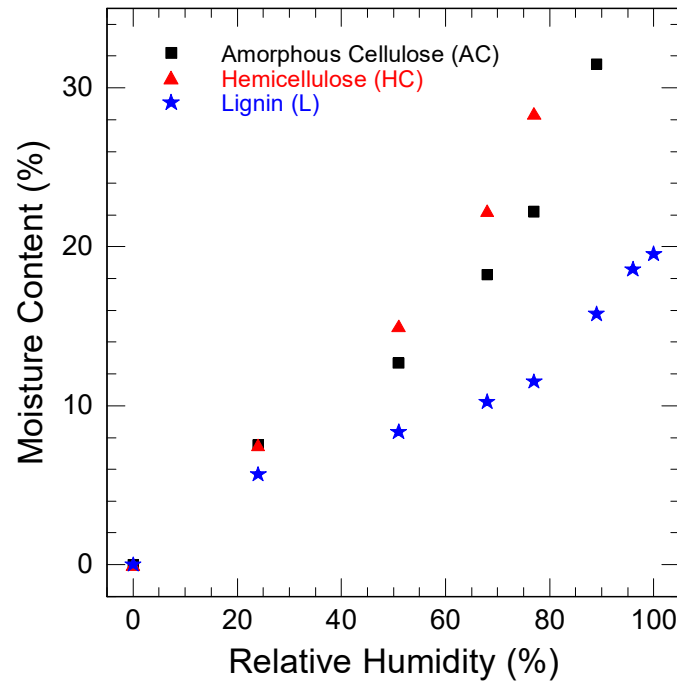


Figure 1.10 Sorption isotherms of isolated wood polymers.⁶³

1.2.1 Moisture-induced swelling

In bulk wood, changes from the FSP to oven dry cause anisotropic dimensional changes of approximately 8%, 4%, and 0.1% in the tangential, radial, and longitudinal directions, respectively^{64,65}. However, these changes are not representative of the swelling of the cell wall probably due to presence of lumina and different cell shapes, whose deformation may also accommodate some of the swelling.⁶⁶ Cell wall thickness and number density of cells also play a role in the total swelling of wood, and studies have shown that latewood cell walls have lower volumetric strain and more isotropic transverse swelling.⁶⁷

At the S2 cell wall level, phase contrast X-ray computed tomography studies have shown that swelling perpendicular to the cell wall thickness is much higher than the parallel swelling, moreover, the overall volumetric strain is larger (~ 24%) than the strain reported at the cellular level.⁶⁸

At the elementary fibril level, scattering studies have revealed that water can enter in the less ordered regions between elementary fibrils.^{25,26,33} At this level the moisture-induced strain from dry to 25%MC is approximately 25%. Despite the high strain at this level, studies have yet to address whether this swelling is isotropic or if it is dependent on the primary plane of the scattering volume.

At the crystallographic level, synchrotron x-ray diffraction measurements have shown that the cellulose crystalline lattice is deformed with increasing MC likely due to an increase in the swelling pressure exerted by the surrounding matrix.⁴⁴ Crystal deformation was anisotropic, and changes were only found in the a and c axis (0.6% and -0.2%, respectively). Based on the previous lateral expansion and longitudinal contraction a total volumetric strain of approximately ~0.42% would be expected.

A summary of how wood swells across length scales is provided in Table 1.4. Here, it is evident that the swelling at the elementary fibril level is much higher than the swelling exerted on the crystalline lattice or the swelling strains reported at the S2 level. However, despite the high level of swelling occurring at the elementary fibril level, most studies have not attempted to make a connection between the swelling at this level and higher length scales.

Table 1.4 Summary of swelling in wood across length scales.

	<i>Direction</i>	<i>Moisture-induced Swelling</i> %	<i>RH or MC range</i>
<i>Bulk</i> ^{64,65}	Tangential	8	0 - 30% MC (0% - FSP)
	Radial	4	
	Longitudinal	0.1	
<i>Cellular - latewood</i> ⁶⁷	Tangential	3.36	25 - 85% RH
	Radial	2.65	
<i>S₂</i> ⁶⁸	Parallel to the cell wall thickness	6.89	25 - 85% RH
	Perpendicular to the cell wall thickness	15 - 23	
<i>Elementary fibril</i> ²⁵	Average tangential-longitudinal plane from earlywood	25	25 - 97% RH (6 - 25% MC)
<i>Cellulose monoclinic crystals</i> ⁴⁴	a axis	0.6	0 - 30% MC (0% - FSP)
	c axis	-0.2	

1.2.2 Moisture-induced polymer dynamics and its implications

The presence of water in wood not only swells the structure, but also increases the mobility of wood polymers^{69–71}, which facilitates transport of ions⁷² and moisture-induced relaxations^{73,74}. The moisture-induced dynamics generally involves the mobility of polymer chains, and the creation of free volume as the polymer chains become more mobile. The increase in free volume has been associated with the mechanical softening observed when polymers pass through the glass transition and change from a glassy to a rubbery state.⁷⁵ Due to the interest in establishing a connection between the moisture-induced dynamics and the bulk-scale properties such as softening and transport, several techniques have been used to gain insight on the moisture-induced phenomena across length scales.

The moisture-induced dynamics between 60 – 80% RH, where Kamke et al.⁷⁴ showed that dielectric relaxation seen in wood cell walls agrees with the moisture-induced glass transition in hemicelluloses (~ 16% MC)⁷³, have been of particular interest. Jakes et al. proposed that once the hemicelluloses pass through the moisture-induced glass transition, they form swollen interconnected regions where ions can diffuse.⁷⁶ X-ray fluorescence studies showing that diffusion of ions only occurs above 75% RH, supports the proposed mechanism for ion transport through wood cell walls. Though these studies showed a connection between transport and the glass transition, no further connection was made between the polymer dynamics and the results.

Experimental studies solely focused on the moisture-induced dynamics of wood and wood polymers have been limited to NMR studies that measured the mobility of cellulose and paper.⁶⁹ Other experimental studies have focused on the properties of the bound water instead, but these studies were only able to estimate the diffusion coefficient of water in the wood cell wall based on water absorption⁷⁷ and NMR spin-spin relaxation studies.⁷⁸ While researchers have been interested in the moisture-induced dynamics of the polymers, most experiments have not been able to provide information. Hence, MD simulations have provided most of the current insight on this topic.

MD simulations on cellulose microfibrils have provided some insight on the state of the adsorbed water, particularly in terms of the water that is loosely bound to the wood polymers in the crystalline-amorphous interface between cellulose microfibrils.¹¹ These studies have shown that as water molecules get adsorbed into the microfibril, they form clusters that increase in size until the maximum moisture content is achieved. According to Kulasinski et al. the increase in porosity and breakage of hydrogen bonds is responsible for the moisture-induced softening of the hydrophilic polymers.¹⁰

More recent MD simulations on the softening of wood polymers⁷⁹ have also shown that when water enters the cell wall, it first occupies all empty space. Then, as water permeates the

hemicelluloses, its fractional free volume increases, whereas in lignin, which has more free space for water molecules to go in, the free volume initially decreases and above 10%MC it increases with further hydration. For both lignin and hemicellulose, the increase in the free volume resulted in softening of the polymers, as revealed by their reduced elastic moduli.⁷⁹

1.3 Needs for research on the moisture-induced swelling

Despite the importance of understanding the nanoscale origins of the moisture-induced swelling for the development of new treatments meant to impart moisture-durability, most studies have focused on the swelling at the cellular scale. While several techniques have been used to study moisture-induced swelling across length scales (Figure 1.11), methods that can probe in situ how the wood cell wall nanostructure changes due to moisture uptake and, moreover, how these changes are affected by chemical treatments, are necessary.

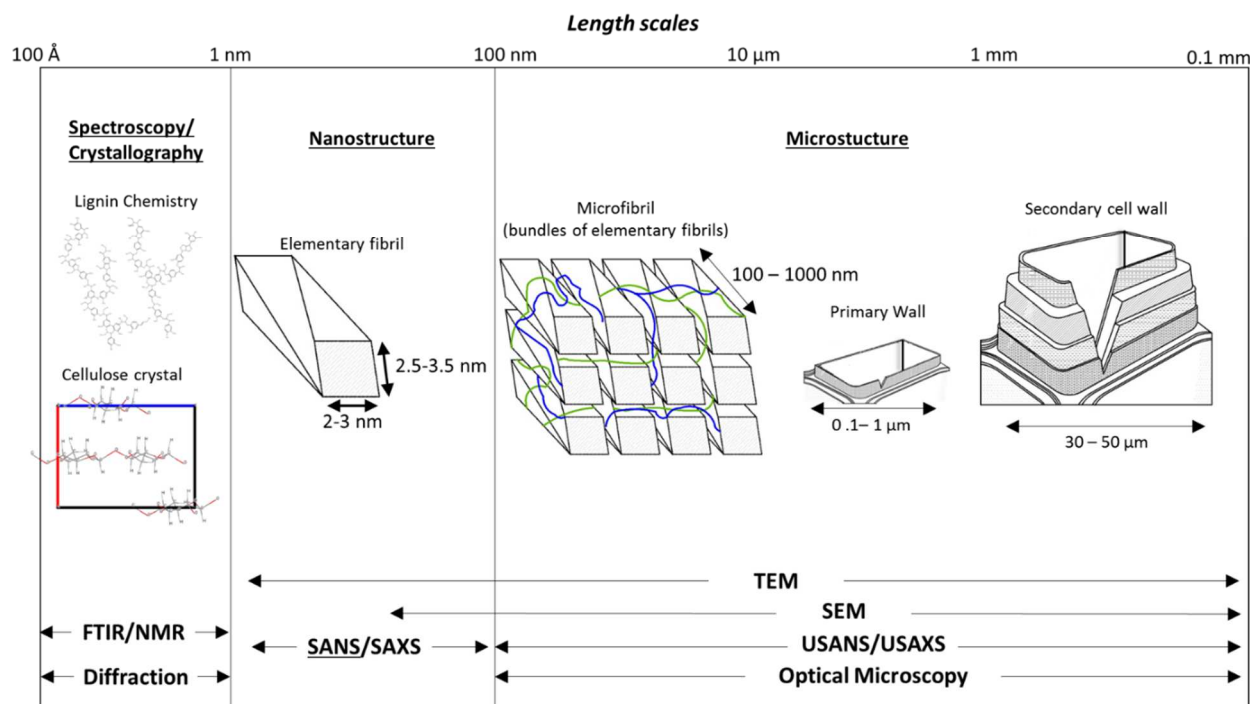


Figure 1.11 Schematic showing the experimental techniques available to study the hierarchy of the wood cell wall. Adapted from Martinez et al.³⁵

The first half of this dissertation focuses on the development and implementation of small angle neutron scattering (SANS) to study the swelling of unmodified and chemically modified cell walls. Here, SANS was selected because it can probe the swelling of the wood nanostructure, particularly at the elementary fibril level, while allowing in situ humidity control. While there are microscopy-based techniques capable of accessing the length scales of interest, these often rely on heavy sample preparation that could affect the wood nanostructure and thus, lead to results that are sample preparation dependent. Furthermore, most of these techniques often require the use of a vacuum to decrease background contributions, and thus would not allow measurements to be performed with in situ humidity control. Despite apparent SANS suitability to study structural changes in the wood cell wall nanostructure,⁸⁰ the effects of sample preparation have not been studied prior to this work, and thus need to be addressed so that SANS can be used to study chemical modification effects. Our SANS work on unmodified wood showed that while sample preparation is not trivial in the analysis of SANS data, there is no primary plane dependence on the measurements of elementary fibril spacing. Furthermore, our results suggested that effective wood protection treatments must target the swelling at the elementary fibril level because it accounts for over half of the transverse swelling of S2 layer.

Our work on chemically modified wood showed that SANS is a suitable method to test the efficacy of wood protection treatments at the elementary fibril level and provide insight on the previously inaccessible nanoscale interactions between chemicals and wood polymers. Our careful SANS studies on treated wood cell walls provided new insights on the nanoscale regions targeted by the treatments as well as the extent of chemical infiltration at the sub-cellular level. Here, it has been shown for the first time that the regions between the elementary fibrils are accessible to low molecular weight chemicals capable of improving the moisture-durability of wood. Likewise, it has also been shown that this infiltration is not the only route available to increase the moisture-durability of forest products. Another possibility is to modify the surroundings of the elementary

fibrils, namely the microfibril matrix to prevent water from entering the water-accessible regions between the elementary fibrils.

1.4 Needs for research on the moisture-induced dynamics

Most recent attempts to gain insight on the molecular-scale origins of moisture adsorption are based solely on computational and modeling methods, thus, experimental techniques that can validate the models presented are still necessary. While numerous spectroscopy techniques have been used in the past to study the dynamics of wood-water interactions, Figure 1.12, in terms of dielectric and nuclear magnetic resonance relaxation times, these techniques are not suitable to probe the time and length scales accessible via simulations. The last part of this dissertation focuses on the development of the experimental methods necessary to validate these models, as well as, the study of the wood-water dynamics that have been previously experimentally inaccessible. Quasielastic neutron scattering (QENS) was selected not only because of its ability to probe the same conditions accessible in simulations, which makes it a valuable tool to validate the recent models proposed by Kulasinski et al., but also because it can probe experimentally wood-water dynamics that have been previously inaccessible. QENS experiments on the moisture-induced swelling of wood were useful in studying the dynamics of water molecules inside the cell wall, as well as quantifying their diffusion mechanism.

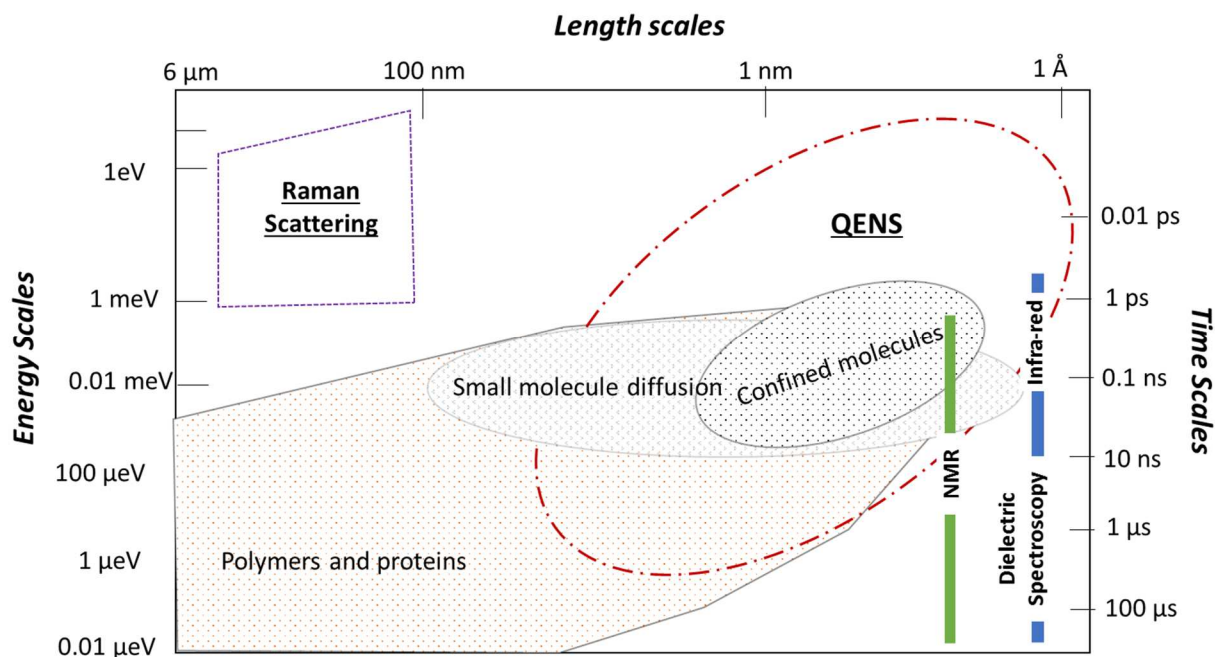


Figure 1.12 Schematic showing the techniques available to study wood-water dynamics. The typical time and length scales of dynamics in polymers and proteins, as well as the diffusion of small molecules are shown for comparison. The length and time scales that QENS can probe, which have been inaccessible experimentally is also shown using a dashed red line.

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Chapter 2. Neutron Scattering of Wood Cell Walls

Experiments that can probe how the moisture content affects both the wood cell wall nanostructure and the dynamics of the absorbed water molecules are needed to improve our understanding of how moisture absorption is related to the changes observed in the bulk properties of wood. Small angle neutron scattering (SANS) and quasielastic neutron scattering (QENS) are experimental techniques suitable to address these needs. However, despite their potential they remain underutilized in lignocellulosic research because forest products are inherently complex due to their hierarchical nature and data analysis is not trivial. In this chapter, a brief background on the techniques will be provided as well as a discussion on the measured quantities that are relevant for forest products research.

2.1 Neutron interaction with wood cell walls

Neutrons are uncharged elementary particles that interact with non-magnetic matter via neutron-nuclei interactions. When a neutron interacts with a single nucleus, the nucleus acts like a point scatterer, and the amplitude of the scattered wave depends on the strength of the neutron-nucleus interaction. The scattering length b is a measure of this strength, and it is determined experimentally via neutron optics.¹ This quantity is an atom property that depends on the isotope but not the proton number Z . The sign of b indicates if the scattered wave was in or out phase with respect to the incident beam. A positive scattering length indicates that the neutron-nucleus interaction was repulsive and the scattered wave was in phase with the incident wave. A negative scattering length indicates that the neutron scattered wave was out of phase with the incident wave. Atoms can also absorb the neutron, which leads to the formation of an excited compound that must decay. Therefore, the neutron-nuclei interaction is best described by a complex potential, where the real component corresponds to b and the imaginary component corresponds to the absorption length b_a . The real component can be further divided into coherent and

incoherent terms. In wood b_a is very small, thus, the absorption is negligible. The average scattering length describing the neutron-nuclei interactions in the sample is known as the coherent scattering length b_c , while the scattering length variance due to spin incoherence is called the incoherent scattering length b_i . Neutron scattering lengths of isotopes relevant for scattering of wood cell walls are listed in Table 2.1.

Table 2.1 Scattering lengths of the atoms found in wood cell walls.²

Atom	Z (proton number)	b_c (1×10^{-12} cm)	b_i (1×10^{-12} cm)	b_a (1×10^{-12} cm)
C-12	12	0.665	0	9.81×10^{-8}
O-16	16	0.580	0	2.78×10^{-9}
N-14	14	0.937	0.20	5.31×10^{-5}
H-1	1	-0.374	2.53	9.24×10^{-6}
H-2	1	0.667	0.404	1.44×10^{-6}

The scattering event can be classified in terms of the energy exchange experienced by the neutron due to the neutron-nuclei and the coherence of the scattered wave. If there is no energy transfer the scattering is elastic, and is described by the scattering triangle shown in Figure 2.1. In this case, the scattered intensity is only measured as a function of the wave vector transfer q ,

$$q = \vec{k}_f - \vec{k}_i = \frac{4\pi}{\lambda} \sin(\theta) \quad \text{eq. 2.2}$$

which can be defined in terms of the incident and scattered wave vectors $\vec{k}_i$ and $\vec{k}_f$, or in terms of the scattering angle θ . Here, $\vec{k}$ is defined $\frac{2\pi}{\lambda}$, where λ is the neutron wavelength.

Elastic Scattering :

$$\vec{k}_f = \vec{k}_i$$

$$E_f = E_i$$

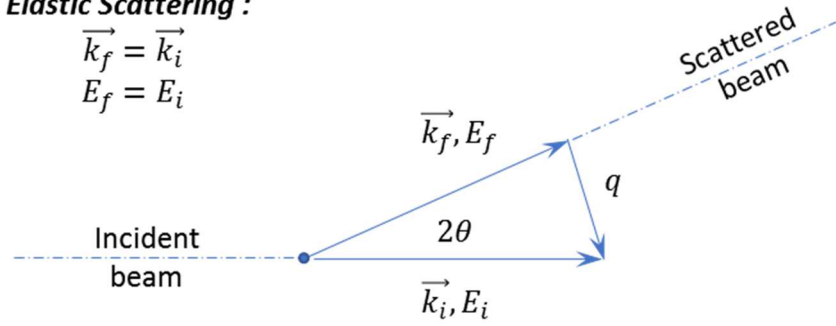


Figure 2.1 Scattering triangle showing the elastic scattering of a single nucleus after interacting with a neutron. The nucleus is shown in the schematic as a point.

Whenever the neutron gains or loses energy, the scattering is inelastic; and is described by the scattering triangle shown in Figure 2.2. A special case of inelastic scattering is quasielastic scattering (QENS), which is the Doppler-broadening effect of the elastic line due to an energy exchange that is almost zero. In this case, the measured scattered intensity has both q and energy dependence. The energy transfer ΔE between the nucleus and the neutron is given by

$$\Delta E = E_i - E_f = \frac{\hbar^2}{2m_n} (k_i^2 - k_f^2) = \hbar\omega \quad \text{eq. 2.3}$$

where E_i and E_f are the incident and scattered neutron energies, k_i and k_f are the magnitude of the incident and scattered wave vectors, m_n is the neutron mass and $\hbar$ is the reduced Planck's constant. In the literature, ΔE is often written as $\hbar\omega$, where ω is the neutron frequency.³

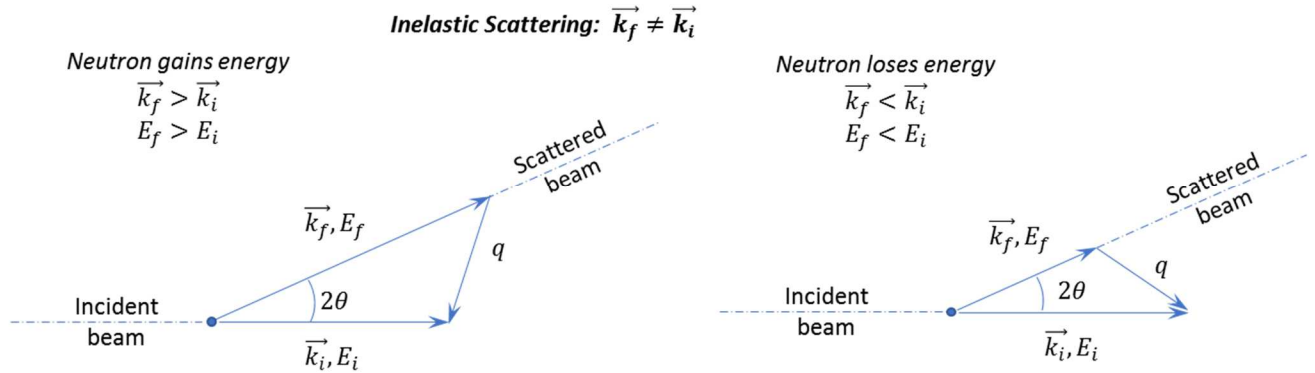


Figure 2.2 Scattering triangle showing the two cases of inelastic scattering of a single nucleus after interacting with a neutron (a) the neutron gains energy and (b) the neutron loses energy. The nucleus is shown in the schematic as a point. Note that by convention the energy transfer is positive when the neutron loses energy.

2.2 Background on Small angle neutron scattering (SANS)

Elastic neutron scattering techniques like small angle neutron scattering (SANS), are generally diffraction-based techniques and well suited to study spatial distributions within the sample. In wood SANS is suitable to probe the cell wall structural changes in the range of 1 nm to over a 100 nm that are caused by moisture uptake as well as chemical infiltration. Using neutrons instead of x-rays offers two main advantages for wood: neutrons scatter more strongly off of light elements and the scattering signal can be further enhanced by deuterating the sample, which is possible to do for wood.

2.2.1 Instrument Geometry

In SANS, a beam of neutrons is focused on a sample and some neutrons are elastically scattered at a scattering angle θ through interactions with atom nuclei. The neutrons scattered at small θ are detected on a 2D detector. The scattered intensity provides information about the size, shape,

and orientation of the 1 – 100 nm structure in the sample. A schematic of a typical SANS experiment is shown in Figure 2.3.

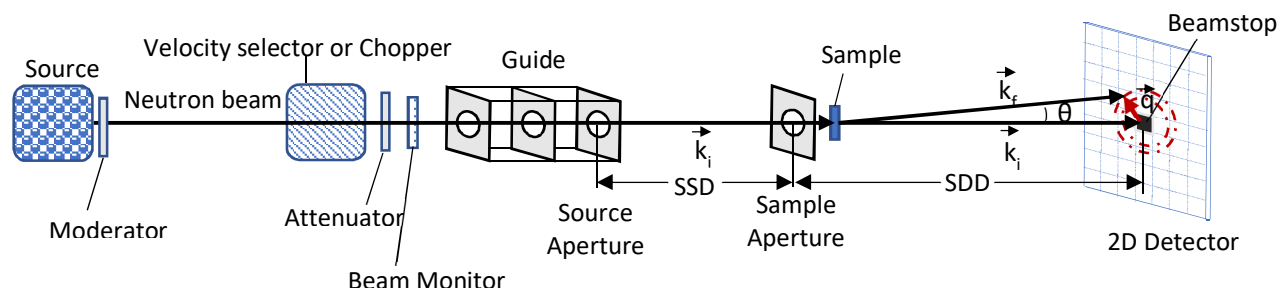


Figure 2.3 Schematic of typical SANS experiment.

The neutron beam is often generated by a nuclear reactor or an accelerator-based pulsed source. In a nuclear reactor, a continuous neutron beam is generated by nuclear fission of a fuel element, such as Uranium 235. Conversely, in an accelerator-based pulsed source, pulses of neutrons are generated by bombarding a heavy element target, such as mercury, with a high-energy proton pulse.

The neutrons generated by either method are fast neutrons (10MeV, 0.000094Å), which are not useful for SANS because their wavelengths are too short to study 1-100 nm structure. Hence, the fast neutrons are cooled down using a moderator to achieve the cool neutrons (0.1-10 meV, 4-30 Å) needed for SANS.

The beam exiting the moderator has a wide range of wavelengths. Instruments with reactor sources use monochromatic incident beams, whose wavelength is selected using a mechanical velocity selector. On the other hand, instruments at pulsed sources use choppers to select wavelength bands. If needed, attenuators are placed after the velocity selector to attenuate the beam intensity and prevent pixel saturation in the detector. The beam then passes through a

beam monitor, which is a low-efficiency gas detector used to quantify the beam strength without attenuating it.

The neutron beam is then directed using neutron-conducting tubes called guides, which are rectangular cross-sectional tubes whose inner surfaces have been coated with a neutron reflective material such as Ni. The guides effectively decrease the source to sample distance (SSD) and preserve the incident beam flux. However, the tradeoff is the guides decrease the monochromaticity of the beam. Because the instrument resolution depends on the beam monochromaticity, using guides decreases the instrument resolution. After the guides, the neutron beam is collimated through the desired sample aperture and passed through a sample.

As the neutrons interact with the sample some will be transmitted through the sample while others will be scattered through interactions with the sample nuclei. The scattered neutrons are detected on a 2D position-sensitive gas detector (LPSD), which consists of an array of cathodic metal tubes containing a neutron-sensitive gas, typically He^3 . As each scattered neutron goes through the detector, it ionizes the gas giving rise to an electric signal detected by the central anodic wire stretched along the tube axis. The electric signal is then used to measure the flux of scattered neutrons in the detector.

2.2.2 Source of contrast in SANS experiment

The strength of the measured signal, as well as, the features that dominate the scattering signal depend on the contrast ΔSLD , which for a two-component system is defined as

$$\Delta\text{SLD}^2 = (\text{SLD}_1 - \text{SLD}_2)^2 \quad \text{eq. 2.4}$$

where SLD_1 and SLD_2 are the scattering length densities of the constituents 1 and 2. An example of a two component system is provided in Figure 2.4.

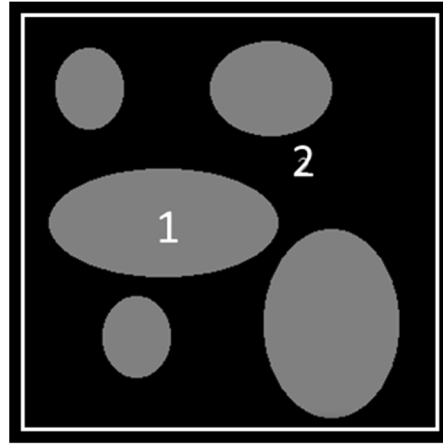


Figure 2.4 Two - component system where each constituent has its own uniform SLD.

The SLD of each constituent x is calculated based on the molecular volume V_m and the scattering lengths of the constituent according to

$$SLD_x = \frac{\sum_j b_j}{V_m} \quad \text{eq. 2.5}$$

Often, V_m is not known and must be calculated based on the molecular mass M , Avogadro's number N_A and the material density ρ , using the following equation:

$$V_m = \frac{N_A M}{\rho} \quad \text{eq. 2.6}$$

Using this equation, the molecular volume of the in-situ wood polymers can be estimated based on the densities and molecular formulas reported in the literature, which were obtained from studies on extracted wood polymers. Then, using equation 2.5, the scattering length densities of the wood cell wall components can be estimated, Table 2.2.

Table 2.2 Estimated scattering length densities of the wood cell wall components. The corresponding densities and molecular formulas are also listed. ⁴

Polymers	Molecular Formula	Density (g/cm ³)	Neutron SLD (10 ¹⁰ cm ⁻²)
Cellulose (crystalline)	C ₆ H ₁₀ O ₅	1.55	1.87
Cellulose amorphous	C ₆ H ₁₀ O ₅	1.48	1.73
Hemicelluloses	C ₆ H ₁₀ O ₅	1.40	1.62
Lignin	C _{9.92} H _{10.68} O _{3.32}	1.46	2.64
D₂O	D ₂ O	1.11	6.33

Because the wood polymers are hygroscopic their SLDs can be changed by soaking the samples in D₂O, which results in increased SLDs due to (1) the deuterium exchange of the H in accessible hydroxyl groups and (2) the addition of D₂O water molecules with a higher SLD than the host polymer. The latter effect can also be achieved by conditioning the samples at a high humidity level if the RH is controlled using D₂O water vapor. The increase in the SLDs of each of the hydrophilic due to the addition of D₂O molecules is shown in Table 2.3.

Table 2.3 Variations in the SLD due to addition of D₂O water molecules. MC refers to the moisture content due to addition of D₂O molecules. The effect of soaking the polymer in D₂O is shown for the pentose groups (amorphous cellulose and hemicellulose), where it was assumed that only three hydroxyl groups were exchanged for deuterioxy. The molecular volume was estimated based on the density of the hydrophilic polymers obtained via MD simulations.⁵

Wood polymer	Chemical Formula	MC %	Molecular Volume (10 ⁻²² cm ³)	SLD (10 ¹⁰ cm ⁻¹)
<i>Amorphous cellulose or Hemicellulose</i>	C ₆ H ₁₀ O ₅	0	1.80	1.75E
	(162 g/mol)	48.6 - 50	3.18	3.63
	C ₆ H ₇ O ₅ D ₃	0	1.87	2.29
<i>Lignin</i>	(168 g/mol)			
	C _{9.92} H _{10.68} O _{3.32}	0	2.05	2.20
	(182.5 g/mol)	19.5	2.59	3.04

Whenever there is more than two SLDs in a material, it can be useful to simplify the system. In solution scattering experiments, this can be done by immersing the sample in a solution that

matches the SLD of at least one of the components in the system. Experiments that rely on the use of solutions with varying ratios of H₂O to D₂O to vary the contrast are known as contrast variation experiments, and the point at which a zero-contrast condition is achieved between two or more components is called the contrast matching point (CMP). A schematic of how the CMP can be used to simplify a three-component system into a two-component system is shown in Figure 2.5. By introducing the zero-contrast condition between the constituents 2 and 3, only the constituent 1 will contribute to the scattering signal.

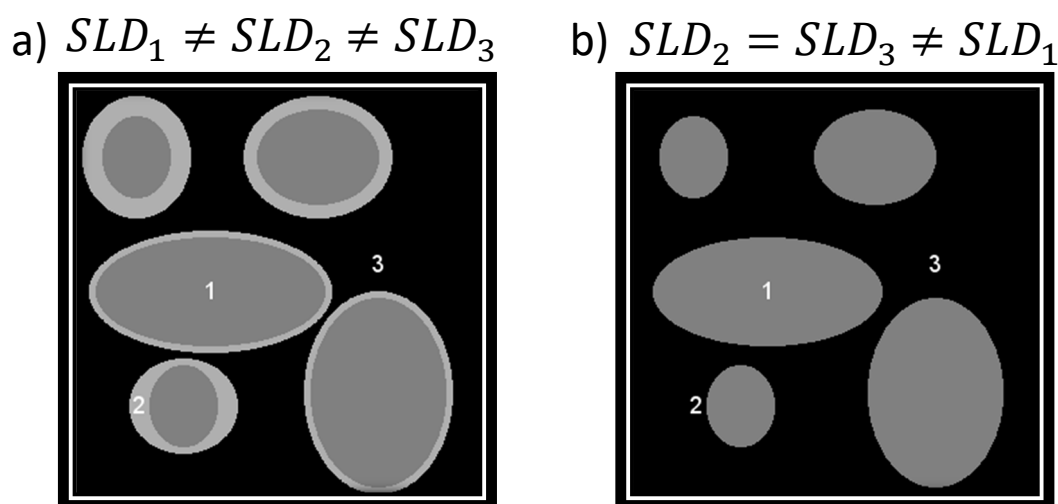


Figure 2.5 Principle of contrast matching experiment. (a) A three-component system that consists of particles with two different SLDs dispersed in a solution with a different SLD. (b) By using a different solution, whose SLD matches the SLD of the constituent 2, the number of visible SLDs is reduced to two.

Generally, to find the CMP of a given material, the SLD of the material is plotted as a function of the volume of D₂O, as shown in Figure 2.6. Note that the SLD used in the example corresponds to the SLD of crystalline cellulose, hence, the SLD does not change with D₂O content because the cellulose does not absorb water, likewise, it is assumed that H/D exchange between the sample and the solution is negligible. Using the Figure 2.6, the CMP can be easily determined by finding the point where the SLD of the material of interest matches the SLD of a D₂O:H₂O

solution, which occurs at the intersection of the two lines shown. The SLD of the solution ($SLD_{D_2O:H_2O}$) varies as a function of the volume content of D_2O ϕ_{D_2O} as follows:

$$SLD_{D_2O:H_2O} = (1 - \phi_{D_2O})SLD_{H_2O} + \phi_{D_2O}SLD_{D_2O} \quad \text{eq. 2.7}$$

Here, SLD_{H_2O} and SLD_{D_2O} are the scattering length density of water and D_2O , respectively. In this case it is assumed that no full H/D exchange is occurring in the mixture.⁶

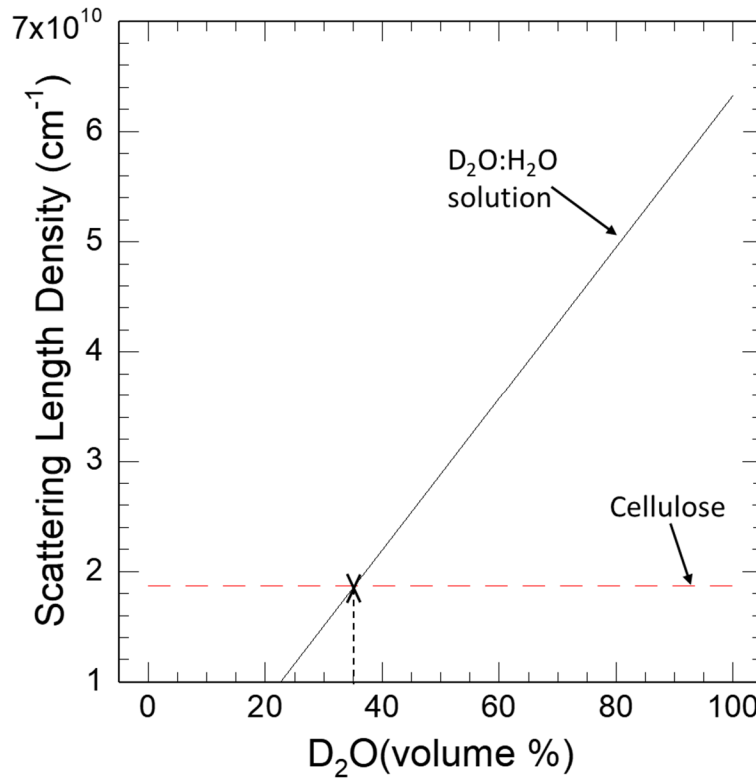


Figure 2.6 Plot of the scattering length density of cellulose as a function of volume content (%) of D_2O in the solution. The CMP of cellulose occurs with a 35% D_2O :65% H_2O solution, which is shown as the D_2O volume % where the two lines intersect each other.

2.2.3 Measured Intensity and the Macroscopic Coherent Cross-section

In SANS, the intensity measured in the detector is the ratio of the scattered flux to the flux of incident neutrons. This ratio is known as the differential scattering cross-section $\frac{d\sigma}{d\Omega}(q)$ and represents the probability that a neutron is scattered into a unit of solid angle ($d\Omega$).

The differential cross section is related to the microscopic cross section σ , which is the cross-sectional area of the nucleus seen by the neutron. This quantity is proportional to the b , and thus, has coherent σ_c , incoherent σ_i and absorption σ_a terms, as shown below.

$$\sigma_c = 4\pi(\sum_j b_{jc})^2 \quad \text{eq. 2.8}$$

$$\sigma_i = 4\pi(\sum_j b_{ji})^2 \quad \text{eq. 2.9}$$

$$\sigma_a = \frac{4\pi}{k}(\sum_j b_a)^2 \quad \text{eq. 2.10}$$

Here, k corresponds to the incident wavevector defined previously. Using these equations, the microscopic scattering cross sections of the wood cell wall components can be calculated and are listed in Table 2.4.

*Table 2.4 Microscopic scattering cross sections of the relevant wood cell wall components. *For the oven dry wood estimate, the oven dry weight of a latewood sample prepared for SANS was used to convert the mass percentage into atomic fractional values.*

Name	Molecular Formula	σ_c $1 \times 10^{-24} \text{ cm}^2$	σ_i $1 \times 10^{-24} \text{ cm}^2$	σ_a $1 \times 10^{-24} \text{ cm}^2$
Water	H ₂ O	0.0393	50.61	0.740
Heavy Water	D ₂ O	5.12	1.39	0.00127
Cellulose	C ₆ H ₁₀ O ₅	0.283	41.4	0.532
Hemicellulose	C ₆ H ₁₀ O ₅	0.283	41.4	0.532
Lignin	C _{9.92} H _{10.68} O _{3.32}	0.449	39.1	0.501
Oven Dry Wood	6%H,50%C,44%O ^{7*}	0.306	41.2	0.0530

However, because the measured cross-section can vary as a function of sample volume⁸ it is useful to define the macroscopic scattering cross section $\frac{d\Sigma(q)}{d\Omega}$ by normalizing $\frac{d\sigma}{d\Omega}$ with respect to the sample volume V illuminated by the beam as follows:

$$\frac{d\Sigma}{d\Omega}(q) = \frac{1}{V} \frac{d\sigma}{d\Omega}(q) \quad \text{eq. 2.11}$$

Note that the experimentally measured scattered intensity $I(q)$ can be equal to $\frac{d\Sigma}{d\Omega}(q)$ but only after it has been corrected for background (I_{bckg}), transmission (T), neutron flux (φ), and detector efficiency (ϵ). These corrections are generally performed as the data is reduced and will be discussed in detail in the data reduction section in the appendix A. Without these corrections, $I(q)$ is only proportional to $\frac{d\Sigma}{d\Omega}(q)$ as shown below.

$$I_{raw} = \varphi A d_t \Delta\Omega \epsilon t T \left(\frac{d\Sigma(q)}{d\Omega} \right) + I_{bckg} \quad \text{eq. 2.12}$$

Here, A is the illuminated sample area, d_t is the sample thickness, $\Delta\Omega$ is the solid angle of the detector cell (also known as pixel), and t is the counting time.

The macroscopic scattering cross-section also has incoherent and coherent contributions. The incoherent contribution is q independent, and only contributes to the background, whereas the coherent term depends on the sample structure. The coherent contribution is often written in terms of the form factor $P(Q)$, which is a function that describes the shape and size of a single particle, the inter-particle structure factor $S(Q)$, the number density of particles $\left(\frac{N}{V}\right)$, and the particle volume V_p .

$$\left(\frac{d\Sigma(q)}{d\Omega} \right)_{coh} = \left(\frac{N}{V} \right)^2 V_p^2 \Delta SLD^2 P(Q) S(Q) \quad \text{eq. 2.13}$$

Here, ΔSLD corresponds to the contrast factor defined previously, which arises from differences in the scattering length densities within the sample. Note that SANS is only susceptible to the contrast, hence, if two samples had different scattering length densities with similar distribution, but same contrast, their SANS profiles would be indistinguishable. An example of this phenomenon, known as Babinet's principle, is shown in Figure 2.7.

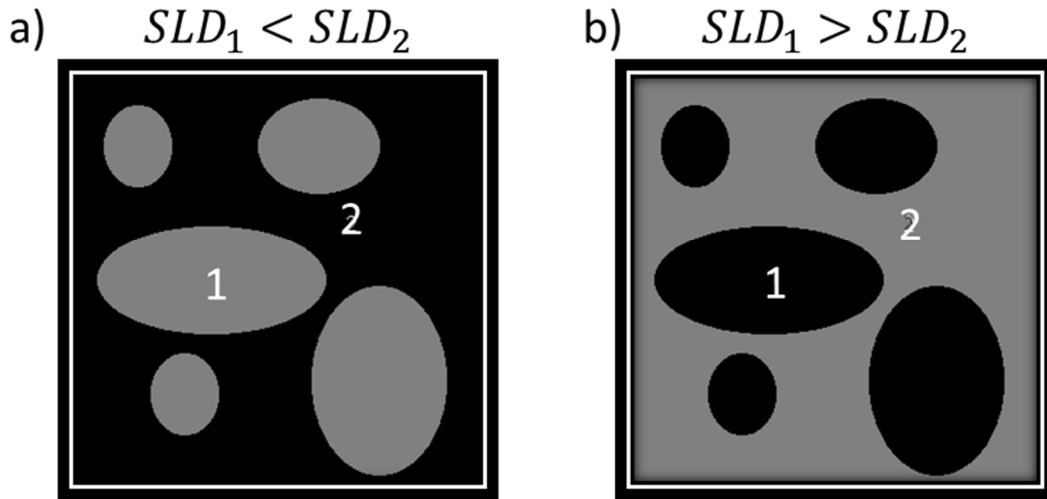


Figure 2.7 Examples of a two-component material whose components have uniform SLDs. Both examples have the same ΔSLD , but in (a) $SLD_1 < SLD_2$ and in (b) $SLD_1 > SLD_2$.

2.2.4 Analysis of SANS data

SANS data can be analyzed to retrieve information about the shape and size of nanostructures as well as their spatial arrangement within the sample. However, analysis of scattering data is not trivial and is generally divided into two main categories: model-independent and modelling based methods. Model-independent analyses can be particularly useful to quantify trends and retrieve general structural information about the scattering features, such as characteristic shapes and sizes, whenever there is little information about the SLD distribution in the sample. Modeling-based approaches, on the other hand, generally require some information about the system, such as the shape of the scattering objects or their SLD distribution. Then, this information can be used to model the scattering profiles using a combination of form factors and structure factors or inverse Fourier transform modeling methods. In this dissertation, the use of model-independent analysis methods was favored because of the minimal assumptions required to analyze the data. The most common model-independent analysis, including power laws, Guinier and small-angle diffraction, will be discussed in detail below.

Power law scattering

Scattering from nano-sized objects generally involves scattering from mass and surface fractals, which includes surfaces of objects whose size cannot be fully resolved in the measured q -range. This results in power law scattering that can expand through one or more decades of q , which is described by the equation

$$I(q) = Bq^{-P} \quad \text{eq. 2.14}$$

where B is the power law prefactor, which typically reflects the contrast and number of objects contributing to the power law, and P is the power law exponent. Scattering from mass fractals such as polymer chains generally results in power law exponents between 1 and 3. The exponents within this range can be further divided based on the dimensionality parameter of the object. For instance, 1D objects such as rods have power law exponents of 1 and 2D objects like sheets or lamellae have exponents equal to 2. Higher power law exponent (between 3 and 4) are attributed to scattering from surface fractals such as smooth surfaces or interfaces, and rough or crumpled surfaces. In this range, the exponent varies depending on the degree of smoothness of the surface. For example, an exponent of 4 indicates the surface is very smooth.

An example of scattering from two objects, namely, aligned rods that form larger lamellar structures, which results in two power law scattering regimes is shown in Figure 2.8. In the figure, scattering with two power law regions is shown. Scattering from rods gives rise to a power law exponent of 1 at the high q , whereas, at low q lamellar structures give rise to a power law exponent of 2. The point where the two power laws intersect each other, indicates a characteristic length scale d in the sample, which is related to q using

$$d = \frac{2\pi}{q} \quad \text{eq. 2.15}$$

In this case, d could be indicative of the length of the rods or of the lamellar width.

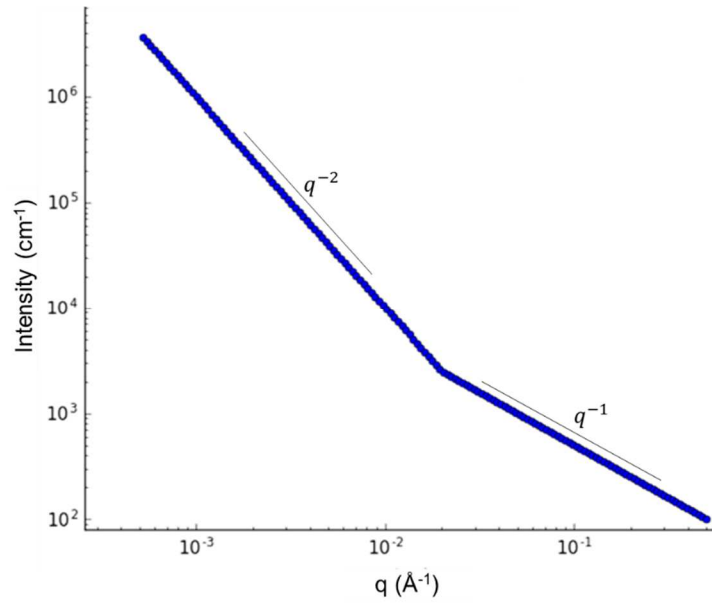


Figure 2.8 Scattering profile from cylinders that form larger lamellar structures. Each scattering shape results in a different power law, thus two power laws are shown. The cross-over between the two power laws is indicative of a characteristic length scale in the sample.

Guinier scattering

Scattering from objects with a regular size can often lead to a Guinier regime, which is typically observed as a shoulder in the scattering profile. However, if there is a high concentration of scatterers this can lead to inter-particle interactions, which can reduce our ability to observe a Guinier regime. If the Guinier region is measurable, the scattering profile can be fitted with the following form factor:

$$I(q) = \frac{G}{q^s} \exp \left[-\frac{q^2 R_g^2}{3-s} \right] \quad \text{eq. 2.16}$$

which allows us to determine the radius of gyration R_g of the objects giving rise to the scattering. Here, G is known as the Guinier prefactor and it accounts for contrast, and number of scattering objects; and s is a dimensionality parameter that changes according to the shape of the scattering objects. For instance, $s=0$ for globular objects or spheres, for rods $s=1$ and for platelets or lamellar

objects $s=2$.^{9,10} An example of a scattering profile with a Guinier region caused by scattering from spheres with a radius of gyration of 6 nm is shown in Figure 2.9.

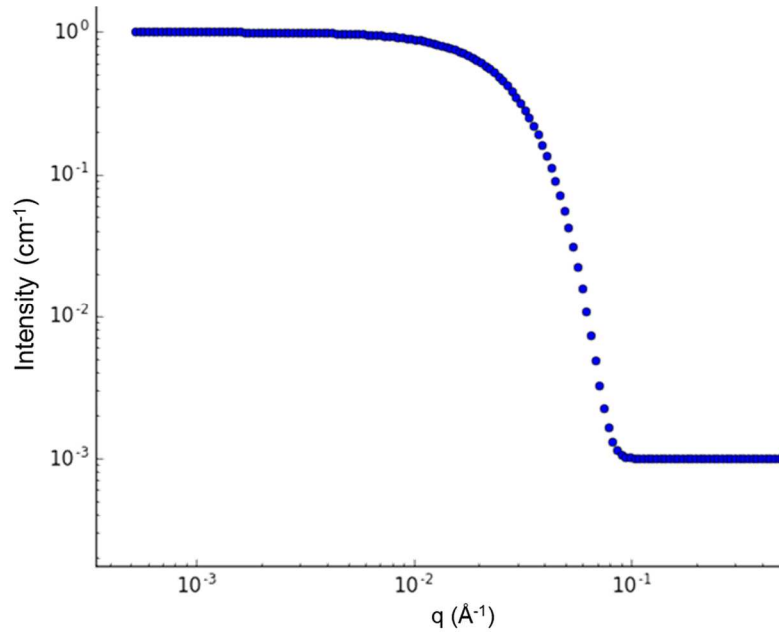


Figure 2.9 Scattering profile from spherical objects with a radius of gyration of 6nm, showing a Guinier region.

Small-angle diffraction

Whenever there is coherence in the spatial arrangement of the nanostructures giving rise to the scattering, this order might give rise to diffraction peaks. These peaks can be analyzed following Bragg's law to obtain the characteristic length scale giving rise to the peak. An example of a small-angle diffraction peak caused by cylinders that are regularly spaced every 5 nm is shown in Figure 2.10. The spacing between the cylinders d is inversely related to the peak position q_0 from the relation

$$d = \frac{2\pi}{q_0} \quad \text{eq. 2.17}$$

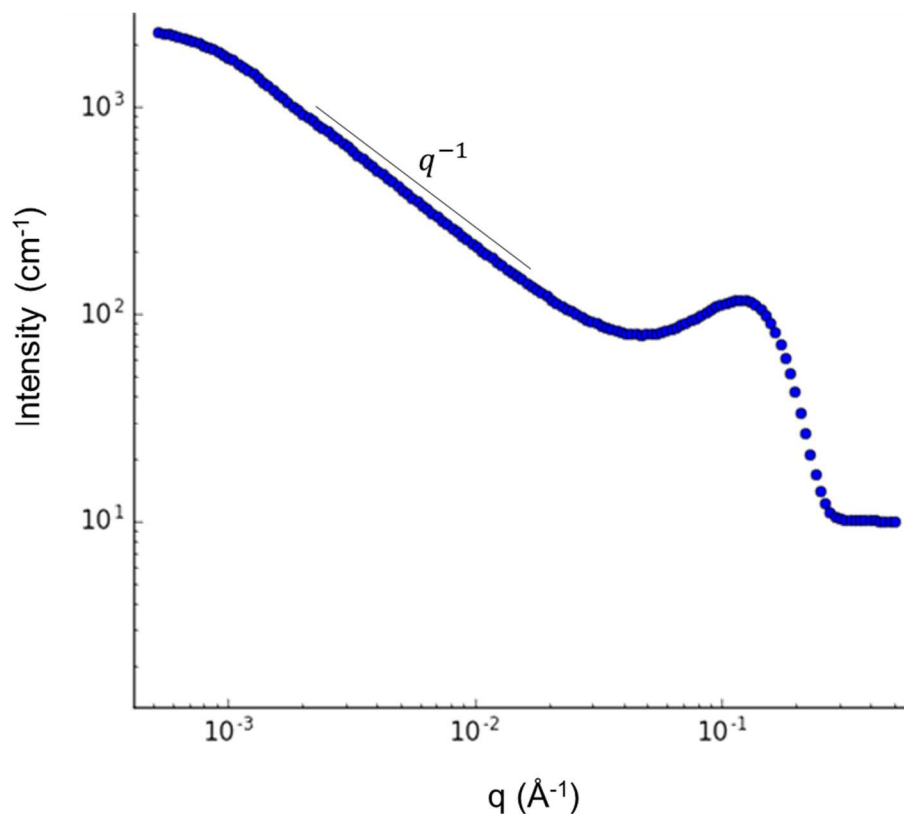


Figure 2.10 Scattering profile from cylinders that are regularly spaced, which gives rise to a diffraction peak. Each cylinder has a radius of 1.5 nm, and a length of 400 nm. The center to center spacing between cylinders is 5 nm.

2.3 Background on Quasielastic Neutron Scattering

Inelastic neutron scattering techniques, like quasielastic neutron scattering (QENS), are spectroscopy techniques that are well suited to study molecular motions whose time scale falls in the pico to nanosecond regime². The measured signal is a function of both the reciprocal vector q and the energy transfer, which is almost zero in QENS. QENS, in particular, is suitable to probe the dynamics of water inside the wood cell walls, particularly in terms of their molecular translational and rotational self-diffusion.^{11,12} Like in elastic measurements, the noise to signal ratio can be tuned by deuterating the samples.

In a QENS experiment, the accessible energy resolution depends on the instrument geometry. Heisenberg uncertainty principle links the energy of a spectrometer to the time scale of the diffusive motion measured. Using this the slowest time scales τ_{slow} accessible based on the energy resolution (δE) can be calculated as

$$\tau_{slow} = \frac{\hbar}{\delta E} \quad \text{eq. 2.18}$$

Similarly, based on the dynamic range of the instrument (E_{max}) the fastest time scales τ_{fast} accessible are given by

$$\tau_{fast} = \frac{\hbar}{E_{max}} \quad \text{eq. 2.19}$$

Table 2.5 lists the current spectrometers' geometries and their resolutions, as well as the dynamic modes that give rise to these energy transfers. The remainder of this section will be focused on the backscattering spectrometer because of its suitability to study the diffusive motions of molecules in confinement and diffusion of small molecules.

Table 2.5 Time scales and length scales accessible via QENS^{11,13}

Spectrometer Type	Time scales		Length scales		Types of dynamics accessible
	Energy transfers (μeV)	Time scale (s)	q ($\text{\AA}^{-1}$)	$d = \frac{2\pi}{q}$ ($\text{\AA}$)	
Direct Geometry time-of-flight	10 - 500	1 – 65 ps	0.05 – 10	0.6 – 125	- H₂O molecules rotation - Phonon dynamics
Backscattering crystal analyzer	3.5 - 250	2.6 ps – 0.2 ns	0.2 – 2.0	3 – 30	- Small molecule diffusion - Dynamics of molecules in confined geometry
Spin - echo	0.005 - 1	0.7ns – 0.1 μ s	0.05 – 3.5	2.5 – 200	- Polymer dynamics - Molecular rheology

In a backscattering spectrometer, a neutron beam with multiple wavelengths is focused on the sample and interacts with the sample nuclei. Then, the scattered neutrons diffract on a crystal analyzer, which is used to determine their wavelength. The analyzed beam is then reflected from the analyzer to the detectors. Figure 2.11 shows a schematic of a typical time of flight (TOF) backscattering spectrometer.

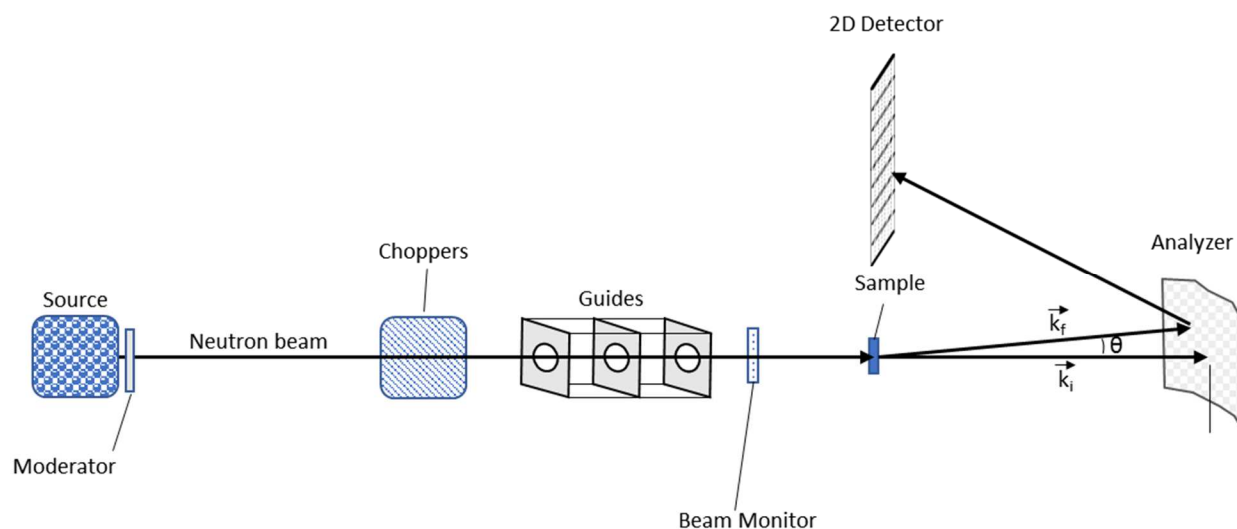


Figure 2.11 Schematic of a typical backscattering spectroscopy instrument

The neutron source controls whether the incident beam is monochromatic. In a TOF backscattering instrument, the incident beam is generated using a spallation neutron source so it is non-monochromatic and the wavelength and energy of the incident neutrons is calculated based on their measured time of flight. In this case, the attainable resolution will depend on the accuracy of the neutron velocity measurement. Conversely, whenever the incident beam is produced with a reactor, the beam is monochromatic and only the scattered neutrons energy needs to be measured.

Choppers or monochromators are used to select the neutrons incident energy. Whenever, choppers are used a wavelength band is delivered instead of a single wavelength; and the center

of the wavelength band corresponds to the d-spacing of the Bragg reflection used in the analyzer. For Silicon 111 crystal analyzers, the incident wavelength that the choppers must select is $6.3\text{\AA}$.¹⁴ Spread in the incident energy is dependent on the monochromator accuracy, and determines the dynamic range accessible in the instrument.

The energy resolution that the crystal can measure is determined either by controlled mosaicity of the crystals or by carefully introducing a spread in the lattice parameter of the crystals. Some of the most common analyzers include pyrolytic graphite, Silicon 111 and Silicon 311.¹³

Once the scattered neutrons have been reflected by the analyzer, they are detected in an array of He^3 LPSD gas detectors. The scattered neutron ionizes the He^3 gas in the detector, which leads to a change in current that is measured. These are the same type of detectors used in SANS instruments, and they are used to resolve the scattering angle.

2.3.1 Measured Intensity and the scattering cross section

In QENS, we measure the double differential scattering cross section $\frac{d^2\sigma}{d\Omega d\omega}$, which allows us to measure both spatial and time correlations between atoms in the sample. Like $\frac{d\sigma}{d\Omega}$, this cross section is also a linear sum of a coherent and an incoherent component. The coherent term depends on the spatial correlation of different atoms as a function of time, thus, it is useful to measure collective motions of atoms. Whereas the incoherent term deals with the spatial correlation of a single atom and is used to measure molecular or atomic self-diffusion.

In these experiments, the quantity of interest is the dynamic structure factor $S(q, \omega)$, which is proportional to $\frac{d^2\sigma}{d\Omega d\omega}$ according to

$$\frac{d^2\sigma}{d\Omega d\omega} = \frac{k_f}{k_i} S(q, \omega) \quad \text{eq. 2.20}$$

Here k_f and k_i are the scattered and incident wave vectors, respectively.

In practice, the dynamic structure factor measured ($S(q, \omega)_{meas}$) is convoluted with the instrument resolution function $R(q, \omega)$, according to

$$S(q, \omega)_{meas} = S(q, \omega)_{sample} \otimes R(q, \omega) \quad eq. 2.21$$

Thus, usually a calibration standard like Vanadium must be used to account for the instrumental broadening due to the instrument resolution.

The measured structure factor has three components: elastic, quasielastic and an energy independent background, and is described by the equation

$$S(q, \omega) = A_0(q)\delta(\omega) + A_1(q)L(\Gamma, \omega) + B(q) \quad eq. 2.22$$

The background B is caused by fast atomic motions, whose time scales are larger than τ_{max} . The elastic component is related to the shape and size of the diffusing entity and is well described by a delta function $\delta(\omega)$. The Lorentzian $L(\Gamma, \omega)$ describes the quasielastic broadening, which is caused by the time scales of the diffusive atomic motions. Here, $A_0(q)$ and $A_1(q)$ correspond to the integrated intensities of each peak. A typical spectrum is shown in Figure 2.12, here the elastic and quasielastic contributions have been highlighted.

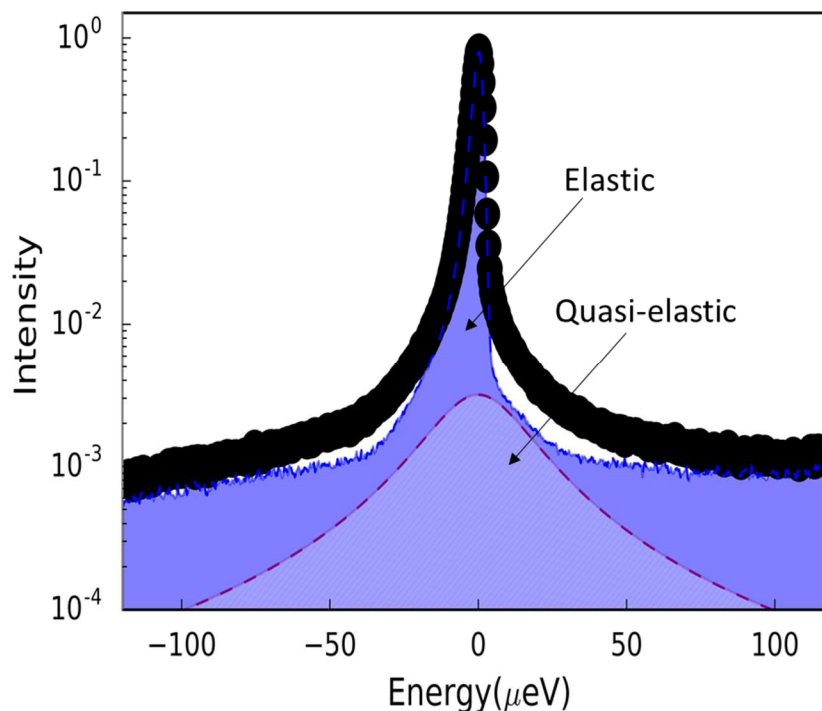


Figure 2.12 Typical QENS spectrum showing the elastic and quasielastic contributions.

2.3.2 QENS analysis

QENS spectra can be analyzed to gain insight on the diffusive motions from the quasielastic broadening of the peak. Moreover, by analyzing the elastic intensity contribution, we can retrieve the elastic incoherent structure factor from which we can learn about the size where diffusion occurs. Similarly, by analyzing the integrated intensities of the elastic and quasielastic contributions we can quantify the static structure factor and the quasielastic incoherent structure factor, respectively. Using the static structure factor, we can learn about the mean square displacements of the molecules that enter the diffusive regime, and from the quasielastic incoherent structure factor we can gain insight from the characteristic sizes and shapes of the diffusive motions. A brief description of how to obtain all these quantities from the measured data will be provided below.

Mean square displacement

Within the limits of the energy resolution set by the analyzer ($\pm\delta E$) no atomic motion can be detected, hence, the QENS signal within these limits ($S_{el}(q)$) has no energy dependence and can be written as

$$S_{el}(q) = \int_{-\Delta\omega}^{\Delta\omega} S(q, \omega) \delta\omega \quad \text{eq. 2.23}$$

In this regime, the intensity is diminished due to the oscillations of atoms about their equilibrium positions. The elastic line intensity is related to the average atomic mean square displacement ($\langle r^2 \rangle$), which describes the amplitude of the atomic motions, according to the Einstein relation

$$\frac{S_{el}(q, T)}{S_{el}(q, T \approx 0K)} \approx \exp\left(-\frac{q^2 \langle r^2 \rangle}{6}\right) \quad \text{eq. 2.24}$$

The experimental mean square displacement includes all atomic motions in the sample. For hydrogenous samples, however, contributions from motions of non-hydrogen atoms to $\langle r^2 \rangle$ are negligible. If $\langle r^2 \rangle$ is not within the length scales accessible during the experiment, no Lorentzian broadening is observed and the measured spectrum is described by a delta function convoluted with the resolution function. Thus, elastic scans, where only $S_{el}(Q)$ is measured, are useful to determine when the atomic motions enter the instrument resolution.

QENS broadening

Each type of diffusive motion is described by a Lorentzian term. While the QENS broadening observed in most systems is well described by a single Lorentzian, hierarchical systems may have more than one type of diffusive motion within the instrument resolution, thus, two or more Lorentzian terms are often necessary to fit the spectra. For instance, in wood cell walls it could be expected to have two dynamic processes: the mobility of wood polymers and the dynamic of absorbed water. However, despite the existence of two dynamic phenomena, the dynamic modes

of polymers are only expected to cause inelastic scattering peaks that occur between 90 and 500 meV.¹⁵ Therefore, only the water dynamics will contribute to the QENS broadening because they are characterized by diffusive motions that are easily probed within the QENS energy range¹⁶. Table 2.6 lists diffusion coefficients of water in different mediums including wood cell wall polymers. Diffusion coefficients for water in individual wood polymers were obtained via simulations where the polymers were held at 95% RH¹⁷, and values for spruce correspond to steady state diffusion measured at 20% MC using the cup method.¹⁸ Note that the diffusive motion of water vapor is too fast for QENS.¹⁹

Table 2.6 Diffusion coefficients of water in different components of wood. All values reported correspond to diffusion at 25°C.

Ion/Molecule	Diffusion Coefficient (cm²/s)
H ₂ O in air ¹⁹	0.3
H ₂ O in H ₂ O ¹⁶	25 × 10 ⁻⁶
H ₂ O in Hemicellulose ¹⁷	5.34 × 10 ⁻⁶
H ₂ O in Amorphous Cellulose ¹⁷	3.46 × 10 ⁻⁶
H ₂ O in Cellulose microfibril ¹⁷	2.532 × 10 ⁻⁶
H ₂ O in spruce along the tangential direction ¹⁸	3.08 × 10 ⁻⁶
H ₂ O in in spruce along the radial direction ¹⁸	3.04 × 10 ⁻⁶

The half width half maximum (Γ) of $L(\Gamma, \omega)$ is related to the diffusion time scale and the shape of $\Gamma(q)$ is determined by the type of the diffusive motion.²⁰ For instance, if a particle can be free to diffuse continuously its diffusive motion is described as free diffusion and $\Gamma(q)$ is described by the following equation

$$\Gamma(q) = Dq^2 \quad \text{eq. 2.25}$$

Here D is the self-diffusion coefficient of the particle.

If the diffusive motion is non-smooth or continuous, and is more accurately described as a stop and go process²⁰, then the diffusion is known as jump diffusion and $\Gamma(q)$ is described by the following equation

$$\Gamma(Q) = \frac{Dq^2}{1+D\tau q^2} \quad \text{eq. 2.26}$$

Here, D also corresponds to the self-diffusion coefficient and τ is known as the residence time, which is the time spent by the particle at a given site before it can diffuse again. The residence time τ depends on the diffusion length l and the diffusion coefficient D according to

$$\tau = \frac{l^2}{6D} \quad \text{eq. 2.27}$$

Characteristic $\Gamma(q)$ curves for both free and jump diffusion models are shown in Figure 2.13

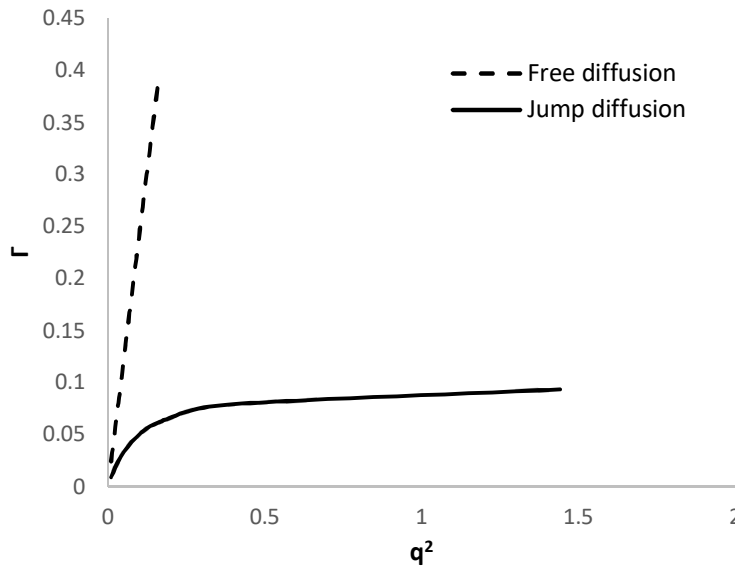


Figure 2.13 Typical curves of $\Gamma(q)$ observed for free and jump diffusion.

Elastic incoherent structure factor

If there are atomic motions in the sample that are measurable within the dynamic range of the instrument, then the geometry of the diffusive motion can be retrieved by analyzing the shape of the elastic incoherent structure factor (EISF). The EISF is the ratio of the elastic intensity to the total measured intensity and is defined using the integrated intensities A_0 and A_1 as

$$EISF(q) = \frac{A_0(q)}{A_0(q) + A_1(q)} \quad \text{eq. 2.28}$$

The EISF is generally fitted to a form factor and the first minimum is usually related to the size of the diffusive particle. For instance, for rotational diffusion of particles, the EISF is described by the form factor of a sphere as follows

$$EISF(q)_{\text{sphere}} = j_0^2(qR) \quad \text{eq. 2.29}$$

Here, R is the radius of the sphere where diffusion occurs and j_0 is the zeroth order Bessel function. The sphere form factor is also commonly used to describe the radius of confinement where molecules can diffuse if details of the diffusion process are limited.²¹

Quasielastic incoherent structure factor

Similar to the EISF, a quantity named the quasielastic incoherent structure factor (QISF) can be defined based on the ratio of the integrated intensities of the quasielastic term to the total intensity measured as shown below.

$$QISF(q) = \frac{A_1(q)}{A_0(q) + A_1(q)} \quad \text{eq. 2.30}$$

The q dependence of the QISF can also be fitted using form factor functions to learn about the shape, size or type of motion giving rise to the quasielastic term.^{22–24} This is particularly useful when there is more than one quasielastic term contributing to the observed quasielastic broadening or when the EISF analysis is inconclusive.

2.4 Summary

The background presented in this chapter is meant to provide sufficient theoretical and practical background on the experimental techniques used in this dissertation, as well as an overview of those quantities that can be obtained using these methods and are relevant when studying the scattering from wood cell walls. A brief description of the analysis methods used in this research was also provided. While the information presented here is also applicable to scattering from other materials, it is important to note that there are scattering events and analysis methods that were not discussed in this chapter and may be relevant in studies involving the scattering from other materials.

2.5 References

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Chapter 3. Moisture-induced swelling of the cell wall ultrastructure

Small angle scattering techniques have been powerful tools to study the cell wall nanostructure for years. Small angle x-ray scattering, in particular, has been used to study the diameter of the elementary fibrils in various types of biomass since the 1950's¹ though it was in the 1990's that a study using SAXS, TEM and WAXS provided some agreement between the techniques in the reported size of the fibrils.² While SAXS of wood has been used to study the hydration dependence of elementary fibril,³ in general SAXS of wood is contrast limited because all wood polymers scatter with similar intensity. Therefore, the structure factor due to the packing of the elementary fibrils is rather subtle, so SAXS is more often used to determine the MFA^{4,5} and/or the diameter of an elementary fibril rather than the distance between elementary fibrils.^{2,6} SANS, on the other hand, has increased contrast because D₂O can be used to introduce high contrast between the wood cell wall polymers and the absorbed D₂O. Hence, SANS has been used to study the packing of the elementary fibrils in various types of biomass at different MCs.⁷⁻¹⁰ In recent years, SANS has become an increasingly popular tool in lignocellulosic research, particularly to study the pretreatment effects on the nanostructure of isolated wood cell wall polymers^{11,12} as well native wood cell walls.¹³⁻¹⁵ Nevertheless, this work is the first attempt to use SANS to study the combined effect of anisotropy and moisture-induced swelling in wood nanostructure across a broad range of length scales.

In this chapter, the moisture-induced swelling across a broad range of length scales (1 – 600 nm) of both intact and partially cut cell walls is discussed. Additionally, the effects of sample preparation will be presented, as well as the effects of scattering from latewood and earlywood cell walls. Most of the analysis presented will focus on the swelling at the elementary fibril, which was found to account for over half of the transverse swelling in the S2 cell wall layer. The SANS data has helped refine a model on the mechanism for the chemical transport through the S2 wood

cell wall layer proposed by Jakes et al.¹⁶, and its implications will be discussed at the end of the chapter.

3.1 Materials and methods

3.1.1 Sample preparation

The effect of cell wall thickness was studied by measuring the SANS patterns from 500- μm -thick earlywood and latewood sections obtained from a single growth ring. Both specimens were obtained only from the tangential-longitudinal plane.

The effect of sample preparation on the scattering pattern was studied by comparing the scattering from an intact 0.5 mm thick latewood section to the scattering from a wiley milled wood sample, where the preferred orientation and arrangement of the cellulose elementary fibrils was obliterated. A vertical column of brass framed sieves with varying sizes was used to separate the ground wood according to their size. For this study, only wood pieces whose size was between 354 and 420 μm were used. The micron sized wood pieces were held between aluminum holders and a mesh.

To study the effect of primary orientation on the SANS data, 500- μm -thick latewood sections of each primary orientation (transverse, radial-longitudinal, and tangential-longitudinal) were prepared from a single growth ring of loblolly pine (*Pinus taeda*) using a razor blade guillotine. Latewood was preferred over earlywood because of its thicker cell walls, which is expected to provide a larger scattering volume.

Additionally, to study the effect of having partially cut cell walls 25- μm -thick tangential-longitudinal and radial-longitudinal sections were prepared using a sled microtome with disposable blades. These thinner sections were stacked to replicate the 0.5 mm scattering length of the thicker intact samples, which required stacking a total of 20 sections.

All samples were secured between two square Al holders (25 mm on a side) with 19 mm circular holes using polyamide tape on both ends, except for the transverse sections. It was observed that the transverse specimens tended to bow out of plane when wetted, hence, to prevent bowing only one end was secured. To fill out the entire area illuminated by the beam, it was necessary to place multiple transverse and radial-longitudinal sections next to each other because each section was only a few mm thick. All specimens were soaked in pure D₂O for 24 hours to improve the noise-to-signal ratio by exchanging the hydroxyl groups in the water-accessible wood polymers to deuterioxyl groups,^{9,13} which decreased their incoherent scattering.

3.1.2 Humidity chamber

To measure samples with in situ humidity control, an RH chamber was designed and built in collaboration with beamline scientists. The custom-built chamber made of aluminum with circular quartz windows (Figure 3.1a) allowed testing of samples conditioned at various humidity levels ranging from 25% to 95% RH. Samples immersed in pure D₂O (100% RH) were also tested using square quartz liquid cells, which were attached to the outside of the chamber with wax. Samples held in the Al holders were mounted inside the RH chamber onto the windows nearest to the detector as shown in Figure 3.1b-c.

Samples were aligned with respect to the neutron beam when mounted. Tangential-longitudinal and radial-longitudinal specimens were placed inside the chamber with longitudinal direction perpendicular to the beam in the vertical direction. The transverse samples were mounted with their tangential direction perpendicular to the beam in the vertical direction.

An L&C Science and Technology (Hialeah, FL, USA) RH-200 relative humidity generator was used to control the humidity in the chamber, which was connected to an external reservoir filled with pure D₂O. Heavy water was used instead of H₂O because minimizing the incoherent scattering from the humidified air is imperative to measure coherent scattering from the samples.

Even low levels of H_2O contamination in the D_2O reservoir can significantly reduce the scattering features of interest, (Appendix A). Moreover, using D_2O increases the contrast between the moisture-swollen wood polymers and the water inaccessible wood polymer.

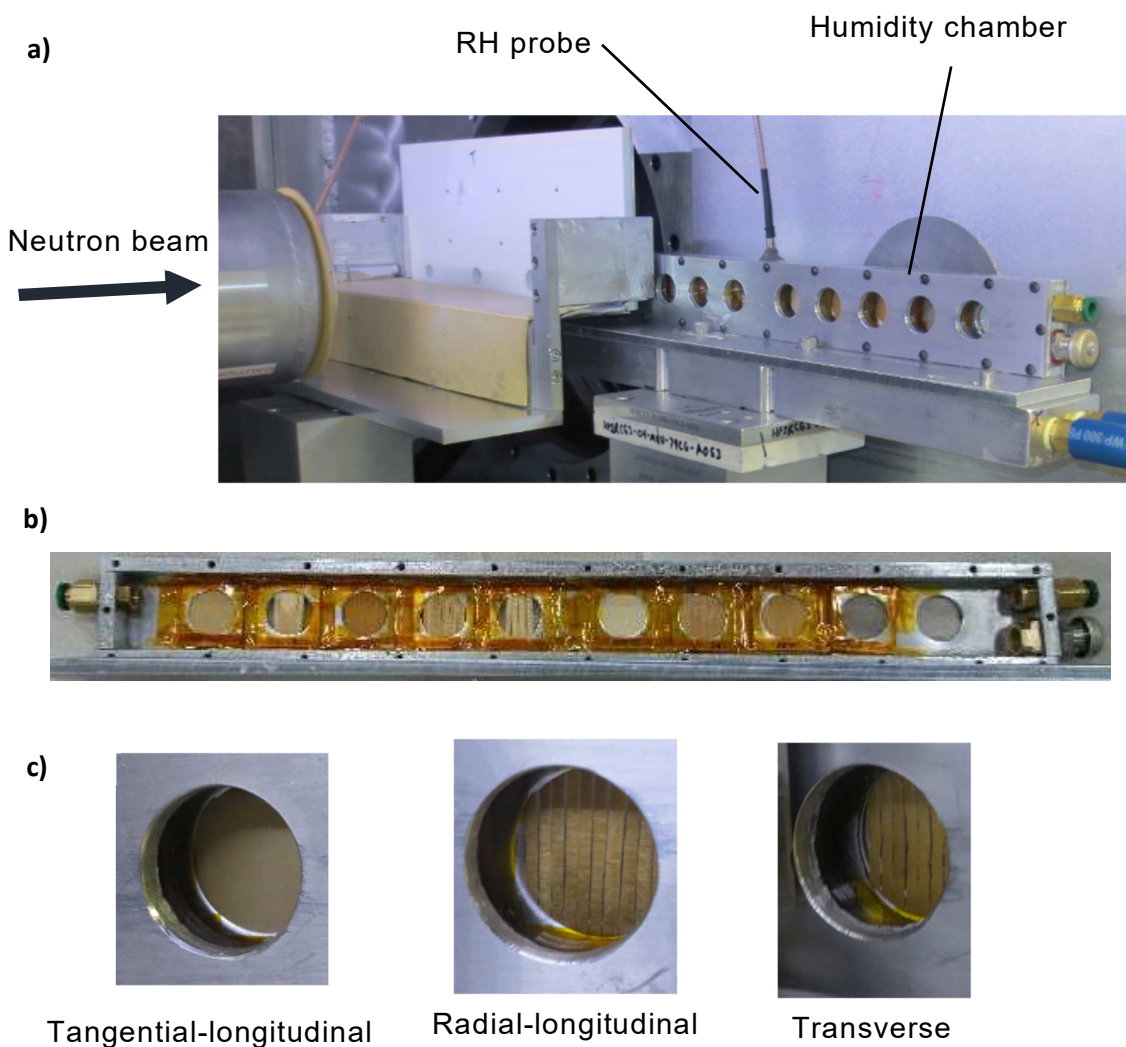


Figure 3.1 (a) Experimental setup used at the Oak Ridge National Laboratory at the HFIR Bio-SANS beamline. (b) RH chamber open to show samples mounted on the back wall. (c) Samples secured in square Al holders as seen through the quartz windows.

3.1.3 Instrument configurations

Two different instruments were used to acquire all the data presented in this chapter. Samples conditioned at different humidity levels were measured at the Bio-SANS instrument¹⁷ at the High Flux Isotope Reactor at Oak Ridge National Laboratory (Oak Ridge, TN, USA), and samples immersed in pure D₂O were measured at the EQ-SANS instrument¹⁸ at the Spallation Neutron Source at Oak Ridge National Laboratory (Oak Ridge, TN, USA). The configuration used at each beam line is described below.

Bio-SANS: Three different instrument configurations were used at Bio-SANS to collect the broadest range of scattering vectors available. To cover the q range of $0.001 \text{ \AA}^{-1} < q < 0.55 \text{ \AA}^{-1}$, a wavelength λ of 6 Å at sample-to-detector (SDD) distances of 0.3 and 6 m, and a λ of 18 Å at a SDD of 14.5 m were used. However, due to the long time required to acquire the full data range (approximately over 3 hours), this range was only collected at 89% RH for all sample orientations and at 45% RH for the tangential-longitudinal sample. Two additional configurations that cover a shorter range were used to acquire data at more humidity level, these measurements only required 40 minutes of exposure. The q range of $0.007 \text{ \AA}^{-1} < q < 0.55 \text{ \AA}^{-1}$ was attained using a wavelength λ of 6 Å, and sample to detector distances of 0.3 and 6 m. Circular aperture diameters of 40 mm and 12 mm for source and sample, respectively, were used for all measurements. A neutron spread $\Delta \lambda / \lambda$ of 0.15 was used for all configurations.

EQ-SANS: Two different instrument configurations were used to attain a broad range of scattering vectors ($0.004 \text{ \AA}^{-1} < q < 0.55 \text{ \AA}^{-1}$). The lower q range ($0.004 \text{ \AA}^{-1} < q < 0.06 \text{ \AA}^{-1}$) was obtained using an SDD of 4 m and a minimum wavelength $\lambda_{\min}$ of 9 Å. The high q range ($0.03 \text{ \AA}^{-1} < q < 0.55 \text{ \AA}^{-1}$) was obtained using SDD of 1.3m and $\lambda_{\min}$ of 2.5 Å. Aperture diameters of 25 mm and 12 mm for source and sample, respectively, was used to collimate the beam.

A 2D (1 x 1 m²) ³He linear position-sensitive detector made up of 192 x 256 pixels was used to record the data at both instruments. All detector images were normalized to the incident beam and monitor counts, corrected for pixel sensitivity, detector dark current and background contributions; and scaled to absolute units. Detector images collected at EQ-SANS were also corrected for time-of-flight. Bio-SANS data were reduced using Igor SPICE routines, and Mantidplot¹⁹ was used for EQ-SANS data.

3.1.4 Anisotropic data reduction

A summary of the anisotropic data reduction used to convert the detector images to 1D scattering profiles will be discussed here. A more detailed explanation is provided in the Appendix B. The first step in reducing the detector images consisted in retrieving the reciprocal space wave transfer vector, which is related to the radial distance from the beam center using

$$q = \frac{4\pi \sin(\theta)}{\lambda} \quad \text{eq. 3.1}$$

where θ is the scattering angle, and λ is the neutron beam wavelength. Then, the 2D SANS data were anisotropically reduced to 1D using the routines provided at the BioSANS and EQ-SANS beamlines. SPICE, a routine written for Igor pro, was used to reduce the data at Bio-SANS. In this tool, the equatorial scattering sector is automatically selected as the azimuthal angle range within 25% of the maximum intensity, and the range within 25% of the minimum intensity corresponds to the meridional scattering sector. Remaining scattering was discarded. MantidPlot¹⁹, an open-source software developed at ORNL, was used to reduce and average the data from EQ-SANS. Here, the sectors were selected manually based on the angular ranges obtained from the SPICE tool. The width for all these sectors was set to 20 degrees. Scattering data from transverse samples was visually inspected with NIKA²⁰, and based on this inspection the sectors were set manually at 20 degrees regardless of the reduction tool used.

Once the data was reduced it was fitted to an empirical model, which allowed us to quantify the differences between the scattering patterns without assuming a specific form factor or a structure factor potential. The selected model included two power laws and a Gaussian peak:

$$I(q) = B_1 q^{-P_1} + B_2 q^{-P_2} + A_G e^{\frac{(q-q_0)^2}{\sigma^2}} + B_3 \quad \text{eq. 3.2}$$

Here, P_1 and P_2 correspond to the power law exponents, B_1 , B_2 and A_G are scaling factors, and q_0 and σ describe the Gaussian peak position and width, and B_3 is the constant background. Data were fitted using the non-linear least square method provided in the Modelling II tool in the Irena package²¹, and the uncertainty of each parameter was determined by calculating the range of values where $\chi_{fit}^2 < 1.05\chi_{critical}^2$.

3.2 Results

3.2.1 Effects of sample source: earlywood vs. latewood

The measured scattering intensity is proportional to the amount of material irradiated by the beam, hence, changes in the cell wall thickness or in the density of the material are expected to affect the overall intensity. Since the cell wall thickness can vary substantially even within a single growth ring from 3 μm thick in earlywood to 6 μm thick in latewood²², it is important to take into account cell wall thickness when selecting the type of wood that will be studied with SANS. Despite its importance, most SAS studies do not take this into account when selecting biomass material. Figure 3.2 shows the effects of sample density on the two-dimensional scattering patterns by comparing the scattering from earlywood and latewood samples obtained from the same growth ring. Here, it can be observed that the scattering from the earlywood sample is not only less intense but this decrease in counts also led to less pronounced features. This trend can be noticeable throughout the entire q range probed. For instance, in the low q range while earlywood still exhibits diamond-shaped scattering, the aligned scattering in the equatorial axis is

not as visible as in the latewood sample. In the high q range, the intensity of the diffraction peak is greatly reduced in the earlywood sample even though the position does not seem to be affected. The higher intensity in the high q region observed in latewood could be attributed to latewood having higher amounts of S2 in the cell wall, which increases the amount of highly aligned elementary fibrils. The trends observed suggest that reducing the total sample volume irradiated can hinder our ability to fully detect the scattering features.

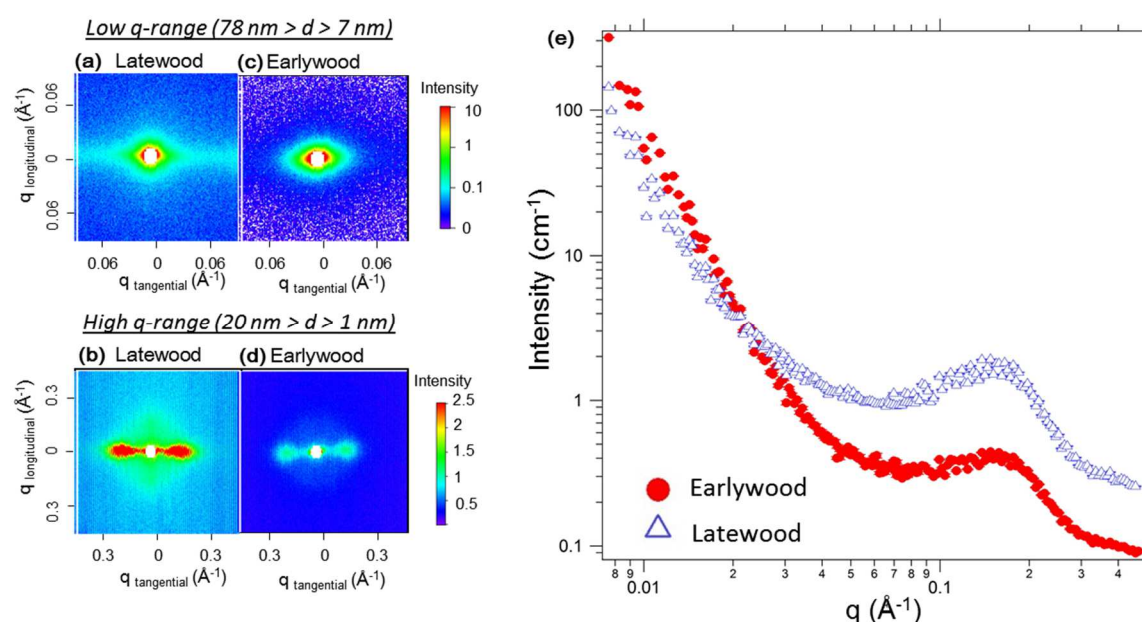


Figure 3.2 Two-dimensional anisotropic scattering patterns obtained from samples conditioned at 89% RH. Patterns correspond to measurements from tangential-longitudinal 0.5mm thick (a-b) latewood and (c-d) earlywood sections. Data was collected at the Bio-SANS instrument at the ORNL-HFIR user facility. (e) 1D scattering profiles of earlywood and latewood conditioned at 89% RH.

3.2.2 Effects of sample preparation

One of the advantages of a neutron scattering experiment is the minimal sample preparation required, which is very advantageous for hierarchical materials. Previous studies on biomass and lignocellulosic materials have often used wood chips or extracted material rather than intact wood cell walls, mostly because these samples were representative of the raw materials used in

biorefineries.^{11,15,23} However, these sample preparation practices can significantly change the scattering features observed because they remove some of the structural order, particularly at higher length scales. Figure 3 shows an example of how Wiley milling can remove all the order that gives rise to anisotropic scattering patterns. The isotropic patterns are observed throughout the entire q -range probe, suggesting that sample preparation must be used carefully because it can effectively remove most of the native arrangement of the elementary fibrils and cellulose microfibrils, thus, limiting our ability to study its swelling or how it is affected by chemical modifications.

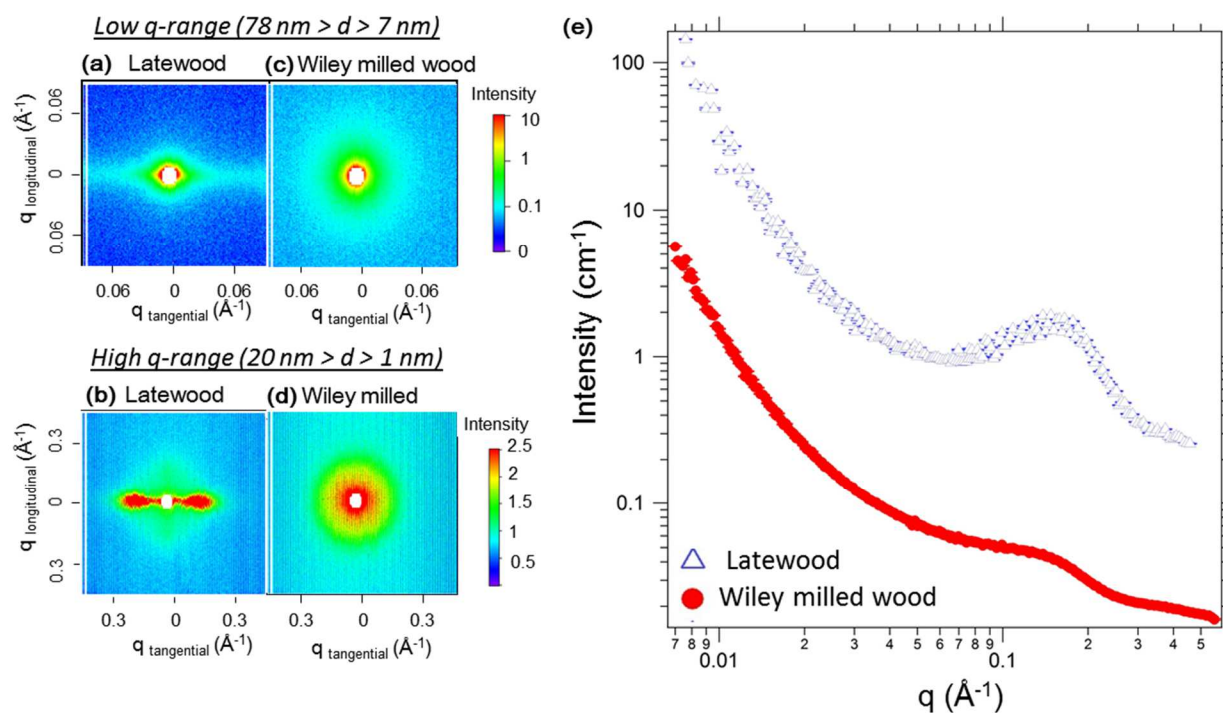


Figure 3.3 Two-dimensional anisotropic scattering patterns obtained from samples conditioned at 89% RH. Patterns correspond to measurements from (a-b) tangential-longitudinal 0.5mm thick latewood and (c-d) ball milled wood that was sieved using a mesh of 420 μm . Data was collected at the Bio-SANS instrument at the ORNL-HFIR user facility.

3.2.3 Anisotropy effects on the moisture-induced swelling

2D SANS patterns obtained from sections prepared from each primary plane, namely, radial-longitudinal (RL), tangential-longitudinal (TL) and transverse (T) show anisotropic scattering features throughout a broad q range, Figure 3.4. Only small differences were observed between scattering patterns from longitudinal samples, while much larger differences were observed between the transverse sample and the other two.

Large structural features ($54 \text{ nm} < d < 600 \text{ nm}$) contributed to the scattering at the lowest q range, Figure 3.4 a, d and g. In the TL and RL samples the lamellar structure in the S2 cell wall layer likely gave rise to the pronounced scattering along the equatorial axis. In the RL sample, the equatorial directional scattering was more pronounced than the TL sample, which suggests that this sample has more pronounced long-range order. This observation agrees with the orientation of the longitudinal tracheids in the RL samples, where the longer side of its rectangular cross-section was oriented parallel to the beam, thus, a larger volume fraction of S2 cell wall lamellae were parallel to the beam. Additionally, distinct meridional directional scattering was observed in RL samples, which can be attributed to the presence of ray tracheids²⁴ whose long axis is aligned perpendicular to the neutron beam in the horizontal direction. These ray tracheids like longitudinal tracheids also have secondary cell wall layers where cellulose microfibrils are organized in lamellar structures that scattered in the meridional direction. Ideally, in the T sample the longitudinal axis of the tracheids would have been parallel to the neutron beam. However, post experiment visual inspection of the transverse sample showed that the longitudinal axis of the tracheids was tilted about 16° with respect to the incident neutron beam. This misalignment led to bidirectional scattering that was enhanced along an axis about 10° from the meridional axis, which suggested that there was a higher volume fraction of S2 cell wall lamellae along this direction. Similar to the RL section, the transverse section had ray cells, whose long axis is

oriented perpendicular to the neutron beam in the horizontal direction. However, their meridional scattering contribution was not observable.

Smaller features ($7 \text{ nm} < d < 82 \text{ nm}$) led to similar anisotropic patterns in the mid q -range, Figure 3.4b, e, and h. At this length scale, cellulose microfibrils were expected to scatter, however, no structural information could be retrieved about them. Lack of structural information could be caused by lack of long-range order in the cellulose microfibrils, weak contrast between the microfibrils and surrounding regions, or scattering too convoluted with contributions from other cell wall features. The subtle differences between the patterns and the lack of diffraction peaks suggested that at this length scale only form factor scattering was observed.

Anisotropic scattering at high q (Figure 3.4c, f, and i) was dominated by scattering from the elementary fibrils, which included a diffraction peak due to contrast between the regions accessible to D_2O and the inaccessible semicrystalline elementary fibril. Diffraction peaks were observed in all patterns; however, the location of the peaks in the detector varied between samples. Only scattering from the elementary fibrils embedded in the longitudinal tracheid cell walls contributed to the diffraction peaks because of their large volume fraction compared to the ray tracheids. RL and TL samples exhibited strong equatorial diffraction peaks, whereas asymmetric peaks were observed in the T sample. The equatorial diffraction peaks have been observed before, and are attributed to the distance between neighboring elementary fibrils.^{9,25} The similarities between the patterns from TL and RL samples indicated that at this length scale the scattering is not dependent on radial or tangential orientations. The asymmetry of the peaks in the T section occurred due to slight misalignment of the elementary fibrils with respect to the incident beam. Asymmetric patterns have also been observed in wide angle x-ray micro diffraction studies of crystalline cellulose from Norwegian spruce and flax fibers whenever the cellulose microfibrils were tilted with respect to the incident beam at an angle higher than the Bragg angle but less than 90° .^{26,27}

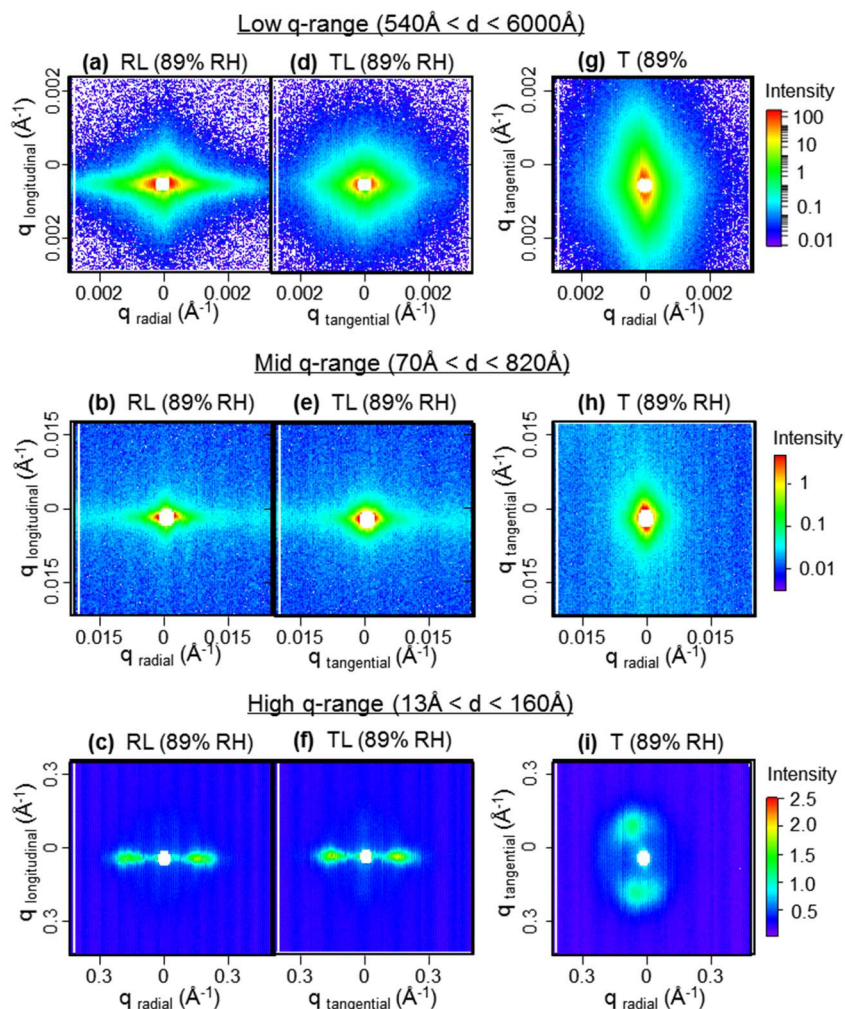


Figure 3.4 Two-dimensional anisotropic scattering patterns of wood from each primary orientation: (a-c) radial-longitudinal, (d-f) tangential-longitudinal and (g – i) transverse. All patterns were obtained at Bio-SANS and samples were held at 89% RH. Low q-range data (a, d and g) were obtained using a sample to detector distance (SDD) of 14.5 m and a wavelength λ of 18 $\text{\AA}$. The mid-q range (b, e and h) and high q range (c, f, and i) data were obtained with SDDs of 6m and 0.3m, respectively, and a λ of 6 $\text{\AA}$

The effects of humidity on the SANS patterns are shown in Figure 3.5. As the RH decreased, the low q diamond shape scattering became more intense due to an increase in contrast at this length scale. At higher q the decrease in the RH led to overall attenuated intensities. This decreased intensity may have contributed to the increased contrast observed at lower q. At the highest q, a shift in the diffraction peak towards higher q was also observed meaning that the spacing between the elementary fibrils decreased with decreasing RH.

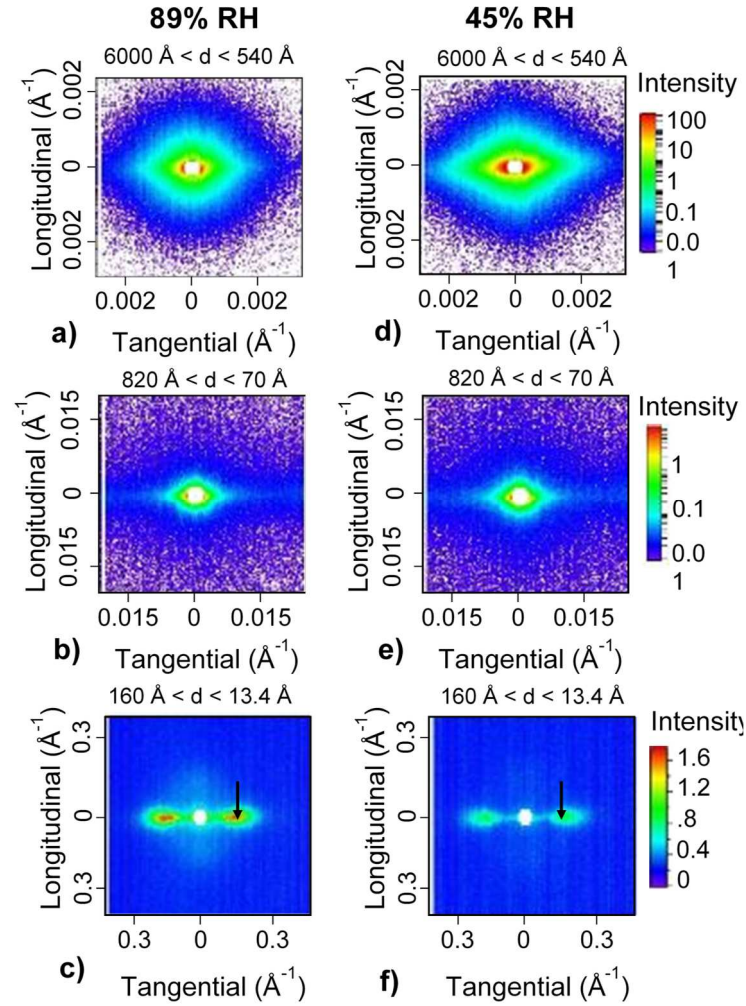


Figure 3.5 Changes of wood anisotropic scattering features as RH decreases. Scattering from tangential-longitudinal samples held at (a-c) 89% RH and (d-f) 45% RH. Data were obtained at Bio-SANS using (a, d) a sample to detector distance of 14.5 m, a wavelength of 18 Å, and a sample to detector distance of (b, e) 6 m or (c, f) 0.3 m with a wavelength of 6 Å. Data were obtained with zero guides.

Data were anisotropically reduced using symmetric sectors for the TL and RL samples, and asymmetric sectors for the T sample, as shown in Figure 3.6. Then, the reduced data were fitted to the empirical model shown in equation 3.2. The fits to the data are shown in Figure 3.6c.

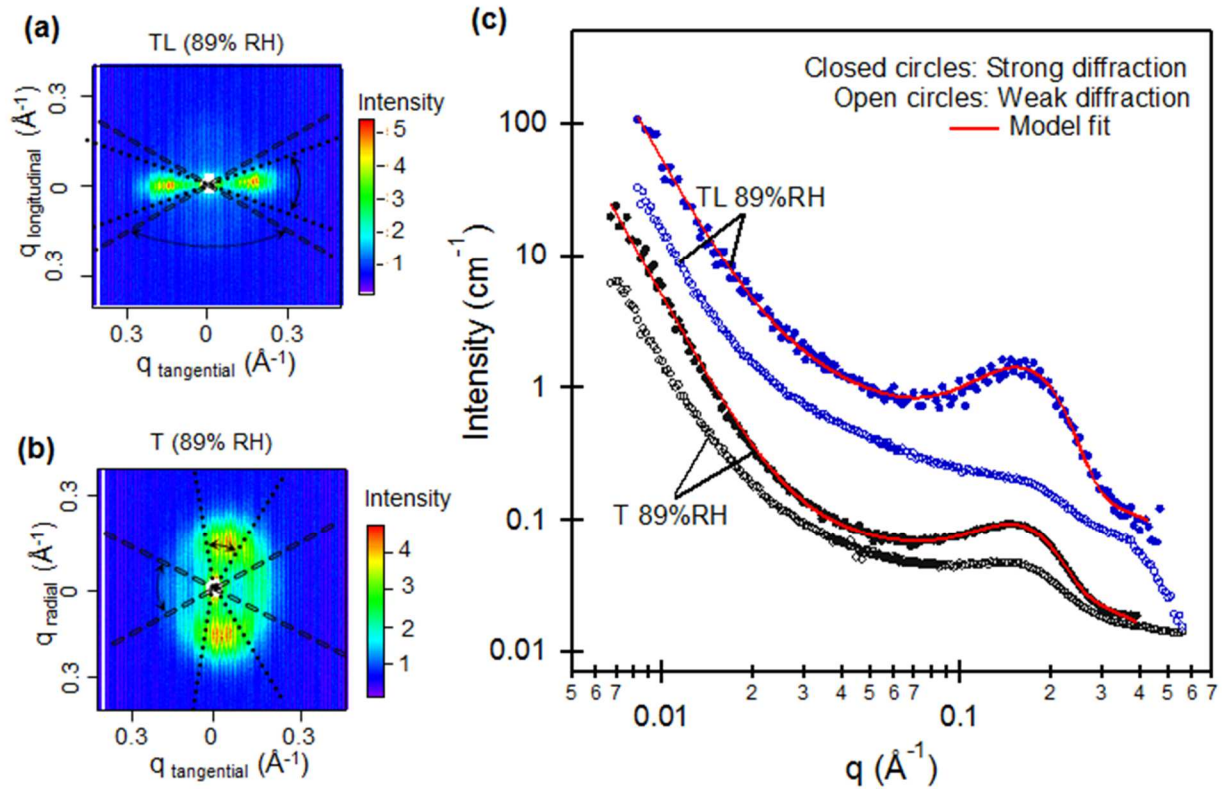


Figure 3.6 SANS patterns from a) a tangential-longitudinal (TL) and b) a transverse (T) sections held at 89% RH showing representative strong diffraction sectors (dotted lines) and weak diffraction sectors (dashed lines) used for anisotropic data reduction. (c) Azimuthally averaged data in absolute units from the strong diffraction sectors (closed circles) and weak diffraction sectors (open circles). The fits of the strong diffraction data to a model $I(q)$ of two power laws and a Gaussian peak are also shown (red line).

In the lowest q -range ($q < 0.01 \text{ \AA}^{-1}$), a power law of $\sim q^{-4}$ was observed for all samples regardless of orientation and RH condition. According to Porod²⁸, a power law of $\sim q^{-4}$ is generally observed at high q , and is caused by scattering from well-defined surfaces of objects whose size contributes to scattering at a lower q . In this case, it can be attributed to scattering from well-defined surfaces from an object whose size is larger than the length scales probed in the experiment, such as the concentric lamellae of microfibrils in the S2 cell wall. Here, the constant power law of -4, which remained unchanged by differences in RH, indicated that the texture of the lamellar surface was smooth regardless of the RH condition. While previous researchers have attributed the low q power law of -4 to the scattering from the empty wood cavities like the lumina,¹³ scattering from

the interfaces between the lamellae in the S2 cell wall layer is the most likely contributor to the low q range probed in this experiment because of its higher contrast and number density.

A mid- q power law of $\sim q^{-1}$ was observed at high humidity levels ($> 80\%$ RH), indicative of scattering from rods, which could correspond to either the elementary fibrils or the elongated swollen regions between them. Below 80% RH, lower power law exponents were observed, which suggested that at lower moisture contents the contrast was too weak or the scattering was too convoluted. In the high q region both strong and weak diffraction sectors showed diffraction peaks. Packing of the elementary fibrils embedded in the S2 cell wall layer gave rise to the peak in the strong sector, whereas, scattering from elementary fibrils in the S1 and S3 layers or the ray cells gave rise to the small peak in the weak sector.

The effect of humidity on the scattering profiles is shown in Figure 3.7a. Both the strong and weak diffraction peaks shifted towards higher q with decreasing RH. However, only the peak in the strong diffraction sector was tracked as a function of humidity because the peak in the weak sector could not be fitted reliably. The trend of the diffraction with RH is consistent with elementary fibrils that are getting more closely packed as RH decreases, Figure 3.7b.

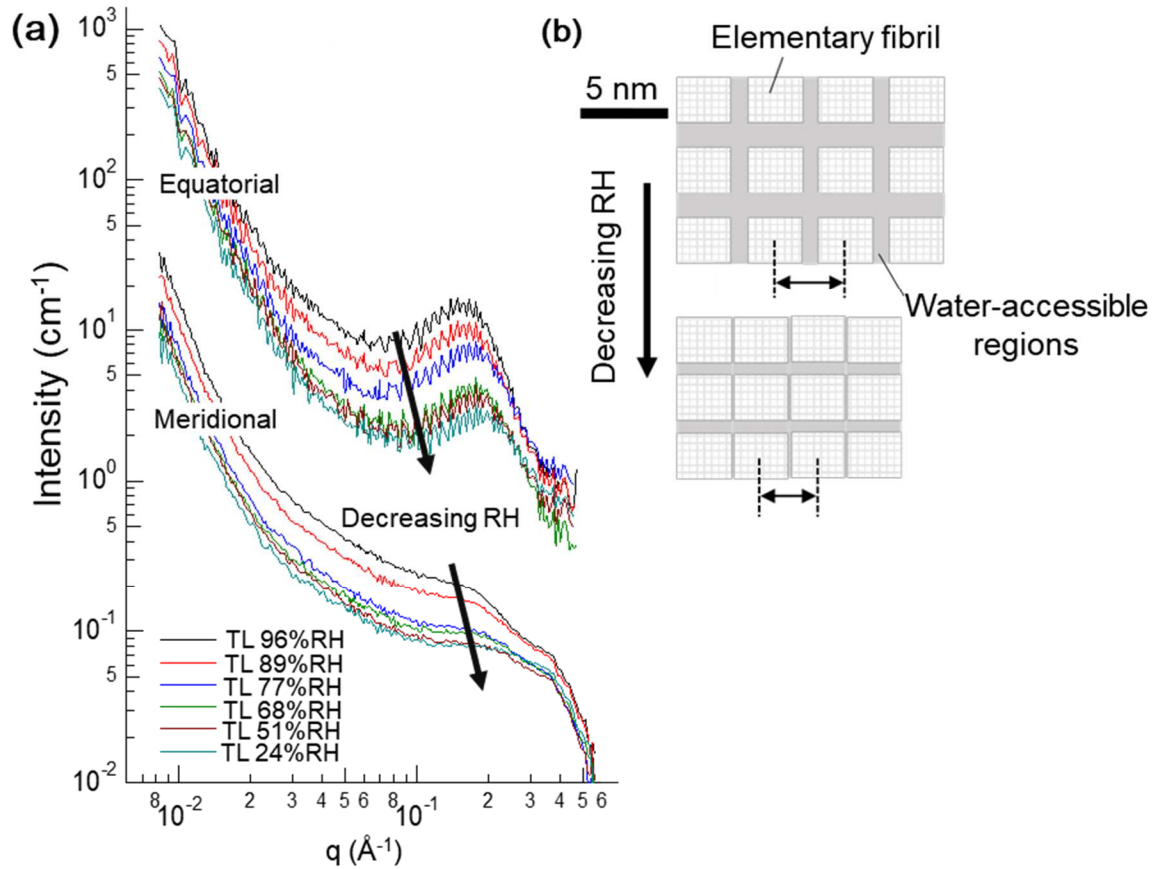


Figure 3.7 Swelling of bundles of elementary fibrils. (a) 1D reduced data from tangential-longitudinal (TL) latewood section obtained from 25% RH to 96% RH during absorption showing how both weak and strong diffraction peaks shift towards higher q as (b) the spacing between elementary fibrils in the microfibrils decreased.

The swelling at the elementary fibril level was tracked by tracking the peak position q_0 , which is related to the elementary fibril spacings d using $q_0 = 2\pi/d$. Interestingly, the swelling of the elementary fibrils follows a similar trend to the MC of bulk wood as a function of RH, Figure 3.8a. In Figure 3.8b, it is shown that the values reported in this study were also found to agree with those reported for earlywood Sitka spruce⁹, despite the difference in wood type. The linear increase in the spacing between 10 and 25% MC also agrees with molecular dynamics simulations.²⁹ However, above 25%MC the spacing reaches a plateau, suggesting that at high MCs either the spacing is no longer proportional to the MC or the maximum amount of water that

can be absorbed at the elementary fibrils was reached. Here the MC of the immersed sample was assumed to be the same as the fiber saturation point, because, by definition, the amount of absorbed water in a sample should correspond to the amount at the FSP.

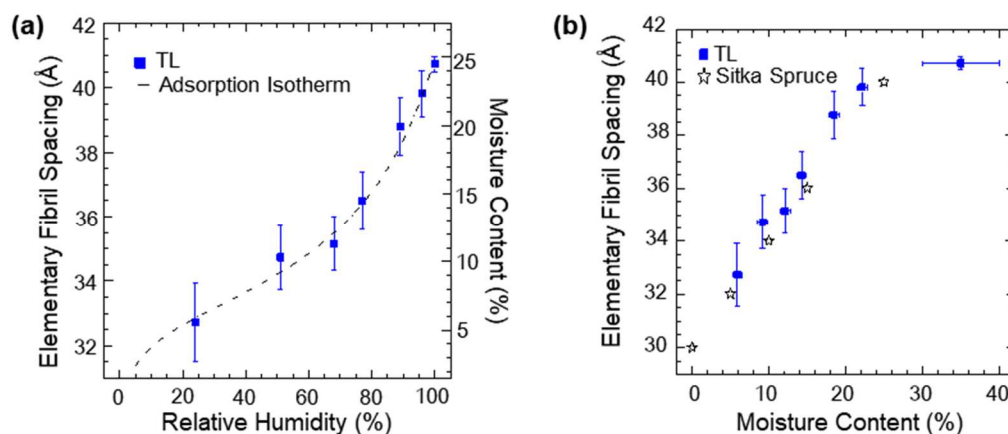


Figure 3.8 Trends in the elementary fibril spacing as function of (a) relative humidity and (b) moisture content. (a) RH dependence of the swelling at the elementary fibril level shows the behavior is similar to Loblolly pine absorption isotherm.³⁰ (b) MC dependence of the swelling at the elementary fibril shows agreement between this study and the values reported on earlywood Sitka spruce.⁹ The MC of the RH conditioned specimens was calculated based on the absorption isotherm while the MC of the immersed samples was estimated as 35% based on the 30-40% MC range for fiber saturation values reported in the literature.³¹⁻³⁴

No differences were observed, within the uncertainty of the measurement, between the elementary fibril spacings measured from each primary orientation, Figure 3.9. This indicates that samples from any wood primary plane can be used to accurately measure the elementary fibril spacing.

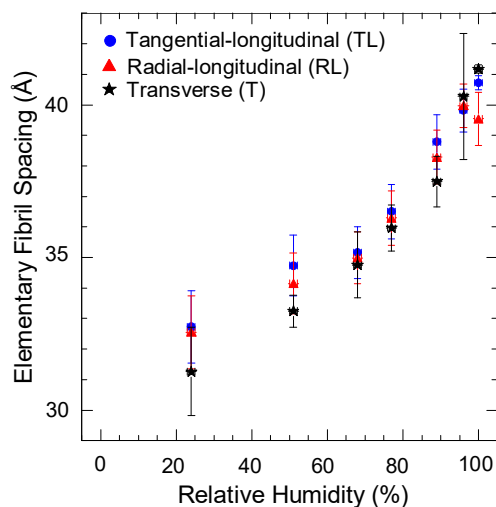


Figure 3.9 Primary plane dependence of the elementary fibril spacing as a function of RH.

3.2.4 Effects of partially cutting wood cell walls

The scattering patterns from intact and partially cut wood cells obtained from the TL orientation are shown in Figure 3.10 to show the effects of cutting the cells on the patterns. At low q , the patterns from the 25- μm thick section stacks were similar to their corresponding 0.5-mm thick sections. This trend was seen for both TL and RL orientations. At high q , the 25- μm thick TL stacked sections scattered more diffusely than the 0.5 mm thick sections (Figure 3.10 c and d), suggesting that there was a slight misalignment in the stacking of the 25- μm thick sections. On the other hand, no differences were observed in the RL sections, suggesting here the stacking was better aligned.

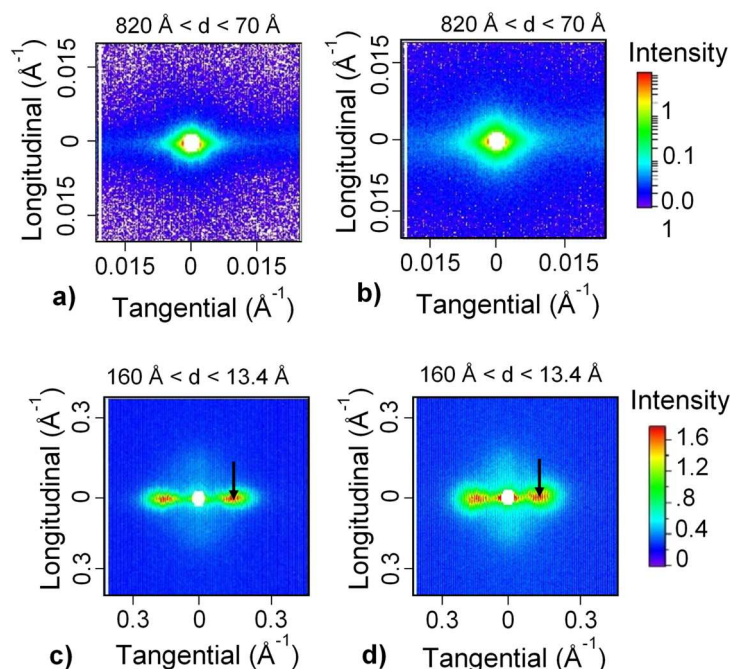


Figure 3.10 Scattering from Tangential-longitudinal samples (a, and c) 0.5 mm thick latewood sections where the wood cell walls are intact (TL) and (b, and d) stack of 25 μ m thick latewood sections where the wood cell walls are partially cut. 2D SANS data was obtained at Bio-SANS using a wavelength of 6 $\text{\AA}$, zero guides, and sample to detector distances of (a, b) 6m, and (c, d) 0.3m.

Once the data was reduced and strong diffraction sectors were fitted, the swelling at the elementary fibril level was tracked. A comparison between the elementary fibril spacing measured from the stacked sections and the 0.5 mm intact samples is shown in Figure 3.11. The stacks only had higher spacings at high humidity conditions (above 90%) and under D_2O immersion, suggesting that swelling of cut cells, like those in wood-adhesive bondlines, only swell more at high moisture content.

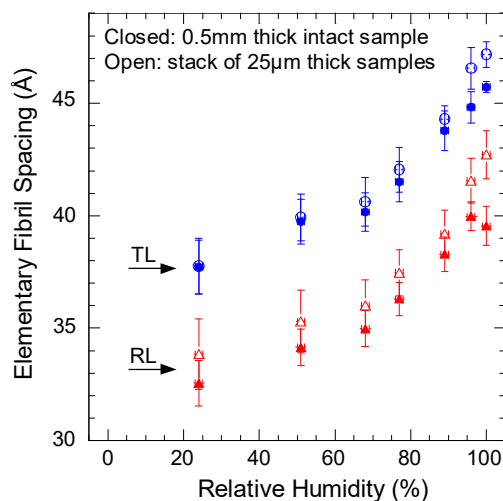


Figure 3.11 Effects of partially cutting the cells in the swelling at the elementary fibril level.

3.3 Discussion

Differences in the anisotropic scattering features show that it is possible to gain insight on the anisotropic nanostructures within the cell wall by carefully controlling sample orientation. It was also shown that sample preparation and alignment with respect to the beam can significantly affect the anisotropy scattering observed. Likewise, our results show that the MC of the samples also affects the scattering and thus, it must be controlled. At the lowest q range measured, scattering from lamellae within the cell wall likely dominated the signal, and the scattering was found to be MC and orientation dependent. Interestingly, while slight misorientations could lead to asymmetry in the high q diffraction peak, it was found that the measurement of the elementary fibril spacing is not orientation dependent.

Dimensional changes driven by changes in moisture content is one of the main durability concerns in forest products. Thus, improving our understanding of how swelling occurs across length scales would accelerate the development and implementation of new treatments that can improve the dimensional stability. While swelling along the longitudinal axis is restrained by the stiff cellulose microfibrils, swelling strains in the transverse plane are much higher and vary across

length scales. The transverse swelling strains from 25 to 85% RH at the cellular, S2 and microfibril levels are listed in Table 3.1. Transverse anisotropic swelling is only observed at the S2 level, while isotropy was observed at the cellular and microfibril levels. The strain measured at the microfibril level is an order of magnitude higher than at the cellular scale, which suggests that the wood structure connecting these lengths accommodates a large amount of the strain.

Table 3.1 Moisture-induced strain in wood at the cellular,³⁵ S2 cell wall,³⁶ and microfibril levels. All strains listed are from latewood in a softwood species

	<i>Direction</i>	<i>Moisture-induced Swelling % (from 25 to 85% RH)</i>
<i>Cellular</i> ³⁵	Tangential	3.4
	Radial	2.7
<i>S2</i> ³⁶	Parallel to the cell wall thickness	2.7 ± 6
	Perpendicular to the cell wall thickness	19.5 ± 6
<i>Microfibril (this study)</i>	<i>Tangential-longitudinal plane:</i>	
	Intact cell walls	17.4 ± 0.8
	Partially-cut cell walls	18.7 ± 0.8
	<i>Radial-longitudinal plane:</i>	
	Intact cell walls	16.5 ± 0.6
	Partially cut cell walls	14.8 ± 0.8
	Transverse plane: intact cell walls	18.7 ± 1

Previously it has been proposed that most of the water is absorbed between the lamellae in the secondary cell walls; and this is why swelling at the S2 level is greatest in the direction normal to the cell wall thickness.^{36,37} Here, I compare swelling at the elementary fibril level with that observed at the S2 level. All calculations will be based on the swelling from 25% to 85% RH listed in Table 3.1. Over the range from 25% to 85% RH (Table 3.1), the S2 swells about 3% and 20% in the parallel and perpendicular directions (with respect to the cell wall thickness). The overall area increase at the S2 level is 23%. While at the elementary fibril level a larger but isotropic

swelling strain of 16 to 19% is observed. Assuming a microfibril rectangular cross-section of 3 by 4 elementary fibrils, this strain causes a total 42% area increase at the microfibril level. Now, if we take into account that around 50% of the S2 area corresponds to cellulose but only 70% of that cellulose is crystalline, then at least 40% of the total area of the S2 transverse plane corresponds to the microfibril.³⁸ Therefore, at least 40% of the S2 experiences a 42% swelling strain, meaning that swelling inside the microfibrils is responsible for up to ~73% of the total transverse swelling strain observed at the S2 level. Note that if the swelling at the elementary fibril was only 16%, the overall area increase in the microfibril level is still about 34%, which corresponds to 55% of the total transverse swelling at the S2. Hence, most of the swelling is originating within the microfibrils and wood protection treatments meant to impart moisture-durability and dimensional instability must address this swelling. A schematic comparing the swelling between the microfibril and S2 levels is shown in Figure 3.12.

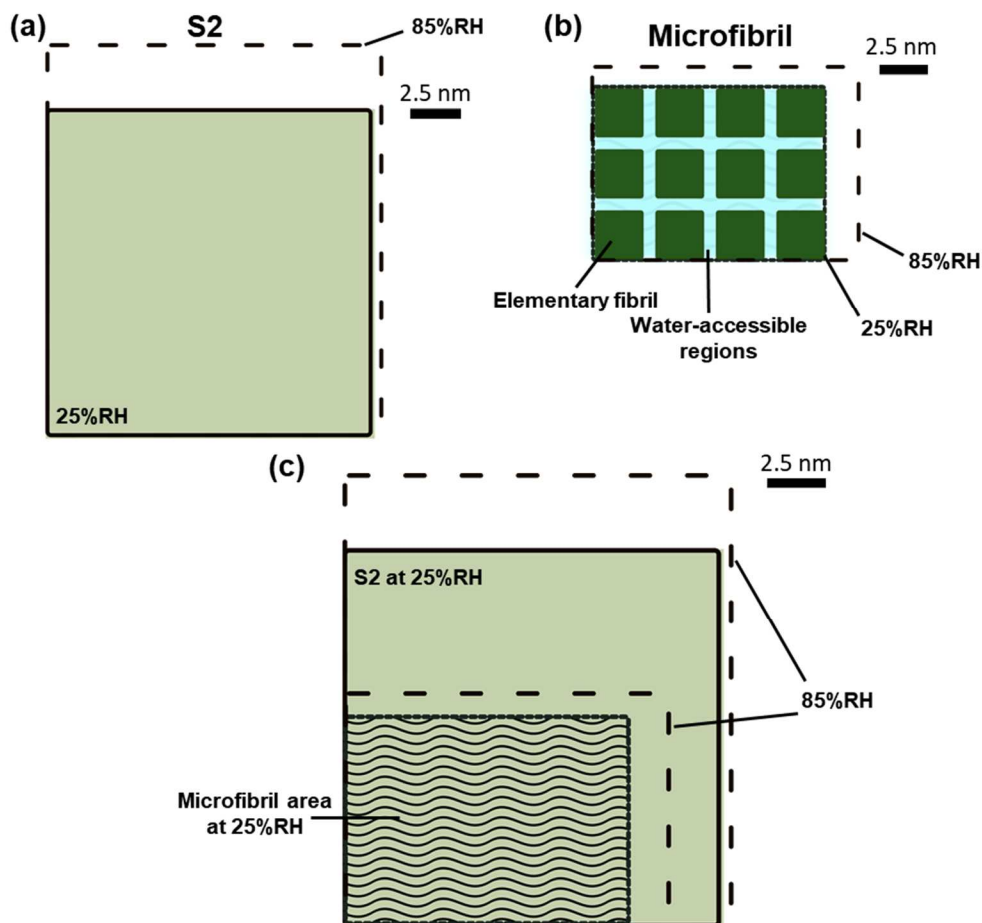


Figure 3.12 Effect of the moisture-induced strain from 25 to 85% RH on the area at the (a) S2 and (b) microfibril levels. (c) Comparison between the moisture-induced swelling strains at the S2 and microfibril levels. The areas at 85% RH are shown using dashed lines.

In this study, the swelling of the microfibrils is not primary orientation dependent and our measurements show no evidence of anisotropy at this level. However, it should be noted that the swelling observed at the S2 level is dependent on the direction with respect to the cell wall thickness, and based on the spot size of the beam used in these measurements and the size of the samples it is not likely that we would be susceptible to this kind of anisotropy. Hence, the swelling measured here is not likely to contribute to the anisotropic swelling observed at the S2 level. The anisotropic swelling at the S2 probably arises from the concentric lamellae. Even if the microfibrils swell anisotropically due to cellulose having crystallographic planes with different

degrees of hydrophilicity,³⁹ the microfibrils would need to be regularly ordered for this contribution to be significant. Nonetheless, it is highly unlikely that such regular arrangement exists at the microfibril and elementary fibril levels. Moreover, both microfibrils and elementary fibrils have a longitudinal twist^{39–41} that would average out any potential anisotropic microfibril swelling contributions to the overall S2 swelling.

Wood surfaces are generally cut via a planer or veneer cutting process before the adhesive is applied in modern forest products like plywood. Hence, in wood-adhesive bondlines the adhesive often interacts with a longitudinally cut wood cell. The specimens made of stacks 25 μm thick sections were meant to mimic these longitudinally cut cells. Additionally, using these sections the role of the S1 and S3 layers on the microfibril swelling was also investigated because in these sections the potential swelling constraint caused by the S1 and S3 layers was removed. Both S1 and S3 layers can withstand large hoop stresses during swelling thanks to the cellulose microfibrils that are embedded at high MFA $>50^\circ$. Hence, it is conceivable that the S1 and S3 layers likely play an important role in constraining the moisture-induced swelling stresses, which have been estimated to be in the order of 90 MPa (from 30 to 100% RH).⁴² However, I found that cut cells only had a higher elementary fibril spacing at the highest humidity and under D₂O immersion (Figure 3.10). Thus, the constraint from the S1 and S3 layers is only important at high levels of moisture-induced strain. Also, the thicker sections (0.5 mm thick) are representative of the swelling of wood cell walls near bondlines up to 90% RH, and can be used in future SANS studies of adhesive infiltration and moisture-induced swelling at the elementary fibril level.

The approximate 1 nm increase in the elementary fibril spacing, Figure 3.8, might be indicative of the formation of diffusion channels for chemical transport between the elementary fibrils. Previously, Jakes and coworkers proposed that the mechanism for transport of chemicals in wood cells was the percolation of softened regions that have undergone a moisture-induced glass transition.¹⁶ However, in the original model the existence of diffusion channels inside the

microfibrils was not considered. These SANS results have informed a refinement of the proposed schematic to include the creation of diffusion channels inside the microfibrils. In the schematic below (Figure 3.13) only the creation of diffusion channels between elementary fibrils is shown. The disordered regions between the elementary fibrils include disordered cellulose chains and possibly the hemicellulose glucomannan.⁴³ Hemicelluloses can be found forming a sheath around the cellulose microfibrils^{44,45} as well as forming networks perpendicular to the microfibrils.⁴⁴ In the proposed mechanism, as moisture is absorbed regions of amorphous cellulose and hemicelluloses become softened as they undergo a moisture-induced glass transition, which is expected to occur in the 11-15% MC range.⁴⁶⁻⁴⁸ As the MC of the sample increases, the rubbery swollen domains form pathways to facilitate transport. While a connection between the MC threshold for ion diffusion and the humidity range of the moisture-induced glass transition of the wood polymers has already been experimentally verified,⁴⁹ a connection between the fibril swelling and the diffusion channel creation or wood degradation has not been confirmed. Nevertheless, if these hypothesized connections are further confirmed SANS could become a valuable tool to further study treatments able to impart decay resistance like acetylation⁵⁰ or heat treatments, as well as develop new improved nontoxic protection treatments.

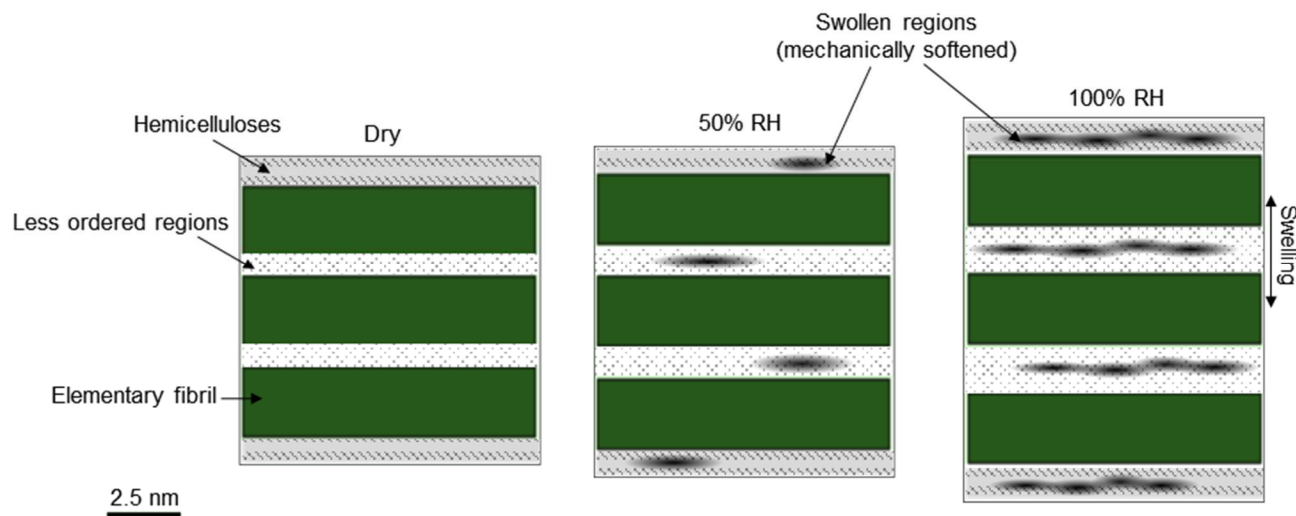


Figure 3.13 Extension of the schematic of the proposed creation of diffusion channels during moisture uptake in the S2 cell wall layer nanostructure. Schematic only focuses on the creation of diffusion channels inside the microfibrils.

3.4 Conclusions

Uniquely anisotropic SANS patterns from each primary orientation provided new insights into the anisotropic structure and moisture-induced swelling of wood. The elementary fibril spacing increased with moisture content up to 25% MC, above which the spacing seemed to saturate. This suggested that before the bulk wood fiber saturation point is reached, the microfibrils have absorbed the maximum amount of water they can hold. Based on simple calculations it was estimated that the swelling at the elementary fibril level accounts for at least 55% of the total transverse swelling of the S2 cell wall layer. Hence, wood protection treatments that mean to improve moisture-related dimensional stability must address swelling at the elementary fibril level. Swelling of elementary fibrils in partially cut cells only differs from intact cells at high humidity conditions, and the elementary fibril spacing measurements showed no primary plane dependence. Finally, it was proposed that as moisture content increases pathways available for transport are created in the regions between the elementary fibrils.

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Chapter 4. Nano-scale structure of chemically modified wood cell walls

To prevent decay and improve its moisture-durability wood is often treated with chemicals that infiltrate and modify wood cell wall structure and properties. Another type of chemical modification occurs due to the application of adhesives to bond wood surfaces in the fabrication of wood composites and timber products. In this chapter, I describe SANS experiments that explore how chemical modifications affect the cell wall ultrastructure. Only two types of modifications will be discussed, namely, adhesive infiltration and acetylation. These were chosen because of their commercial relevance and their potential to represent different interactions with the wood polymers (Figure 4.1).

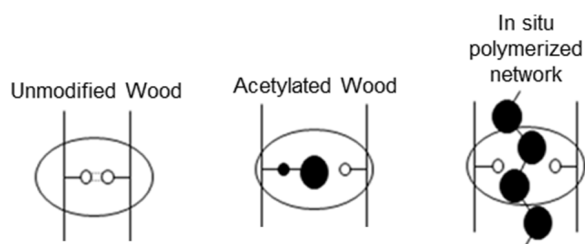


Figure 4.1 Schematic of molecular-scale interactions for different chemical modifications proposed by Norimoto.² In this schematic the vertical lines represent lignocellulosic polymers and unfilled circles are “free” hydroxyl groups. Filled small circles represent hydroxyl groups that were used in the formation of covalent bonds and large filled circles correspond to chemicals that have infiltrated the wood ultrastructure. A change in spacing between the vertical lines implies swelling or bulking due to chemical infiltration into the cell wall.

Most manufactured construction building materials, including plywood and strandboard, are made of wood components held together by an adhesive, such as a thermosetting resin. Phenol formaldehyde (PF), one of the most moisture-durable wood adhesives¹, is thought to be able to polymerize in situ and form an interpenetrating polymer network (IPN). Moreover its bulk modifications can even increase the overall moisture-durability of bulk wood.² The amount of moisture durability attained has been quantified using the anti-shrink swelling efficiency (ASE)

$$ASE = \frac{S_{mod} - S_0}{S_0} * 100 \quad eq. 4.1$$

Here S_{mod} and S_0 are the volumetric swelling coefficients of the treated and the untreated wood, respectively. In both cases, the volumetric swelling coefficient (S) is measured as the amount of moisture-induced volumetric swelling with respect to the oven dry volume as shown below.

$$S = \frac{V(RH \text{ or } MC) - V_{OD}}{V_{OD}} \quad eq. 4.2$$

where $V(RH \text{ or } MC)$ is the wood swollen volume at a specific RH or MC, and V_{OD} is the oven dry volume. Using the ASE, it has been shown that infiltration with either low and high molecular weight PF can result in a significant reduction of the moisture-induced swelling even at low weight percentage gains (WPG) as evidenced by the high ASE values reported, Table 4.1.³

Table 4.1 Effect of low and high molecular weight (MW) PF infiltration in the anti-shrink efficiency (ASE%).³

	WPG %	Anti-shrink efficiency (ASE, %)
Low MW PF	3.6	70.6
	11.3	74.7
	23	86.1
High MW PF	4.1	50.3
	5.8	56.8
	16.5	66.5

Since PF was empirically developed over decades, the mechanisms behind its durability are not well understood. For instance, while it is known that PF with low molecular weight can infiltrate the cell wall, its molecular-scale interactions with the lignocellulosic polymers are still an active area of research.⁴⁻⁶ Moreover, it is widely debated whether or not phenol formaldehyde is

chemically bonded with the lignocellulosic polymers, though recent studies using NMR have proposed that two competing process might occur while PF cures inside the cell wall: lignin degradation and formation of methylene bridges between lignin and PF.^{7,8} Another adhesive that is believed to form an IPN while it cures inside the cell wall is polymeric methylene diphenyl diisocyanate (pMDI).⁹ While pMDI is not as commercially successful as PF, they share some similarities. Like PF, pMDI can also infiltrate into the cell wall structure and its presence in the cell wall can reduce the molecular mobility of lignin and hemicelluloses carbons.¹⁰ The reactivity between pMDI and the wood polymers depends on the molecular size of the pMDI as well as the MC of the cell wall at the start of the reaction. For instance, when dry wood is modified using a low molecular weight pMDI model compound, namely phenyl isocyanate (PI), it is observed that the PI can react with lignin side chains and hydroxyl groups in the hemicelluloses to form carbamates.¹¹ Whereas if a pMDI model compound with a higher molecular weight is used or if the MC of the wood to be treated is higher, the reaction leads to the formation of urethane or biuret structures and the reactivity between the pMDI and lignin is reduced.^{12,13} Here, SANS studies on the moisture-induced swelling of cell walls infiltrated with these two adhesives will be discussed, as well as the extent of adhesive infiltration.

In outdoor applications where moisture is a concern, like window jambs, siding, and decking, wood is often treated to improve its durability. An increasingly popular treatment known to impart decay and moisture resistance is acetylation. In this process, bulk wood is soaked in acetic anhydride to obtain acetylated wood where a portion of the wood polymer hydroxyl groups have been substituted with acetyl groups, Figure 4.2. According to Norimoto¹⁴, this substitution improves decay resistance by initially bulking the cell wall and reducing the number of water-accessible hydroxyl groups. High ASE values for acetylated wood are shown in Table 4.2.



Figure 4.2 Schematic showing acetylation process including reactants and products.

Table 4.2 Effects of acetylation on the swelling coefficient and anti-shrink efficiency (ASE%). Both S and ASE were calculated by measuring the volumetric dimensions of the samples after oven drying and soaking them in water.¹⁵

Acetylation WPG	Swelling coefficient (S1, %)	Anti-shrink efficiency (ASE, %)
0	13.8	--
16.3	5.1	63
22.5	4.1	70.3

The weight percentage gain (WPG) of acetyls is known to play a role in the efficacy of the treatment, and decay resistance is only attained at high levels of acetylation, typically above 20% WPG. However, the distribution of acetyls in the cell wall is still debated. Studies on the acetylation of extracted material have shown that all wood polymers except for crystalline wood cellulose can react with acetic anhydride, and thus become acetylated. However, wood polymers have different levels of reactivity with acetic anhydride, thus, the distribution of acetyls in the cell wall is expected to vary depending on the level of acetylation. For instance, at low levels (~10%WPG) lignin is expected to be fully acetylated because of its high reactivity whereas, only a fraction of the less reactive hemicelluloses would be acetylated.¹⁶ X-ray diffractions studies on cellulose from algae, tunicin and bacteria have shown that acetylation can react with cellulose crystals but only on their surface,¹⁷ which results in cellulose crystals with diminished lateral coherence.¹⁸ While previous studies have focused on the effects of acetylation on the individual wood polymers, here, I use SANS to study how acetylation decorates the wood polymers inside of the intact cell wall to gain insight into how it improves decay and moisture resistance.

4.1 Methods

First, tangential-longitudinal latewood samples 25 x 25 x 0.5 mm thick, were cut from a single growth ring following the procedures described in chapter 3. Then these samples were chemically modified via acetylation or adhesive infiltration as described below. Both hydrogenated and deuterated chemicals were used to modify the samples. Prior to any chemical modification, latewood samples were vacuum dried for 30 minutes to remove moisture.

4.1.1 Acetylation

First, vacuum dried samples were submerged in acetic anhydride using glass stoppers, and heated in an oil bath held at a temperature between 130°C to 140°C for at least 15 minutes to achieve 10% weight gain due to acetylation. Longer heating times were required to achieve higher acetylation levels. Once the samples were cooled in a water bath, these were removed from the acetic anhydride. The remaining acetic anhydride was removed by drying the samples in an oven at 120°C until a constant weight was obtained. The weight percentage gains (WPG) achieved for the samples used in this study are listed in Table 4.3. Samples with deuterated acetyls were obtained using the same method outlined above but deuterated acetic anhydride (D6, 98%) from Cambridge Isotopes was used instead.

Table 4.3 Weight gains due to acetylation for the samples used in this study.

<i>Acetylation Level</i>	<i>Weight Percentage Gain (WPG, %)</i>	
	Hydrogenated	Deuterated
<i>Low</i>	10.8	10.5
<i>Mid</i>	17.5	17.4
<i>High</i>	21.1	22.4

4.1.2 Phenol-formaldehyde infiltration

Low molecular weight phenol formaldehyde was synthesized according to Laborie's methods.¹⁹ To synthesize the resin phenol crystals were mixed with, 37% formaldehyde solution and sodium hydroxide solution in P:F:NaOH molar ratios of 1:2:0.2. Then, the reaction flask was immersed in an oil bath and heated at 80°C for 40 minutes. The vacuum dried samples were immersed in the PF resin for 24 hours using glass stoppers. The samples were removed from the resin, and the excess PF was wiped off. Residual PF was removed from the lumina by vacuum treating the samples for 30 minutes. Then, the samples were heat treated at 170°C in an oil bath to cure the PF. A total weight gain of 27% was measured for the sample infiltrated with hydrogenated PF (H-PF).

Samples infiltrated with deuterated phenol formaldehyde were obtained using the exact same method as for hydrogenated PF but deuterated chemicals were used to synthesize the resin. The following deuterium-labeled reactants were used: d-phenol crystals, 20% d-formaldehyde in D₂O and 40% sodium deuterioxide in D₂O. All chemicals were purchased from Cambridge Isotopes, except for acetic anhydride, which was purchased from Sigma Aldrich. All reagents were used as received. The total weight gain due to infiltration was measured, and a total weight gain of 29% was measured for the sample infiltrated with deuterated PF (D-PF).

4.1.3 pMDI infiltration

Using a chamber housed with saturated sodium chloride the latewood sample was equilibrated to 14% MC. The pMDI adhesive used was Lupranate M-20S (BASF Corporation, Mount Olive, NJ). The specimen was removed from humidity-controlled chamber and weighed, and added to a 20 mL Teflon beak, which was then placed in a 50 mL wide-mouth glass flask with removable multi-port top. The pMDI resin (approx. 5 mL) was then added to the 20 mL Teflon flask (approx. 2:1 NCO:OH molar ratio) until the latewood specimen was completely submerged. The multi-port

top was placed so that two ports were open; one port was used as an inlet of inert gas and the other as an outlet. The entire flask assembly was heated for 2 hours using an oil bath set at 160 °C. The reaction was carried out under a purge of N₂. After the reaction, the specimen was dried at 45 °C overnight under vacuum. The weight gain of the reacted latewood, was approx. 45% (on an oven-dry basis).

4.1.4 Small angle neutron scattering experiments

In-situ humidity controlled experiments: SANS experiments were performed on loblolly pine latewood sections (25 mm wide × 25 mm long × 0.5 mm thick) that had been chemically modified with hydrogenated chemicals. The samples were first soaked overnight in D₂O (99.98% from Sigma Aldrich), and then placed inside the humidity chamber. The relative humidity (RH) was controlled during the experiments using this chamber and an RH generator²⁰. First, the samples were air dried for 12 hours using the RH chamber, held at 0% RH. Then, the experiments were performed in adsorption from 0% to 100% RH at EQ-SANS, and from 25% to 100% RH at Bio-SANS.

Contrast Variation experiments: SANS experiments were also performed on samples that were chemically modified with deuterated chemicals to further elucidate the structure of the chemical modification. To further elucidate the effects of the chemical modification on the structure, the samples were immersed in a solution of 35:65 D₂O:H₂O, which corresponds to the cellulose contrast matching point (CMP) so that there was no scattering contributions from the wood polymers. To measure SANS from the samples while immersed in the solution, the samples were placed in titanium holders with quartz windows. The holders were screw sealed, and the 0.5 mm thick cavity inside the holder was filled with the 35:65 D₂O:H₂O solution using a syringe via cell port on the top of the holder. All air bubbles were removed before the samples were placed in the translation stage at the beamline. By measuring the samples while immersed in the CMP

the scattering from the wood polymers was removed and the scattering from deuterated chemicals was enhanced.

Instrument configuration at EQ-SANS

A broad range of scattering vectors ($0.004 \text{ \AA}^{-1} < q < 0.55 \text{ \AA}^{-1}$) was attained using two different instrument configurations. The lower q range was available ($0.004 \text{ \AA}^{-1} < q < 0.06 \text{ \AA}^{-1}$) with a sample to detector distance (SDD) of 4 m and a minimum wavelength $\lambda_{\min}$ of 9 Å. Whereas, the high q range ($0.03 \text{ \AA}^{-1} < q < 0.55 \text{ \AA}^{-1}$) was available with a SDD of 1.3 m and $\lambda_{\min}$ of 2.5 Å. Data were collected using a source aperture of 25 mm and a sample aperture of 12 mm. Then, scattered neutrons were detected using a 2D ($1 \times 1 \text{ m}^2$) ^3He position-sensitive detector made up of 192×256 pixels. Recorded scattering data were reduced and corrected for dark current, sample transmission, detector sensitivity and background using MantidPlot²¹.

Instrument configuration at Bio-SANS

To cover the entire q range available: $0.0009 \text{ \AA}^{-1} < q < 0.55 \text{ \AA}^{-1}$, SDD distances of 0.3 and 6 m combined with a wavelength $\lambda = 6 \text{ \AA}$, and 14.5 m at $\lambda = 18 \text{ \AA}$ were used. Only the contrast variation experiments were collected using this full range.

To study the effects of acetylation on the moisture-induced swelling, data were collected at various humidity levels over a shorter q -range ($0.007 \text{ \AA}^{-1} < q < 0.55 \text{ \AA}^{-1}$). Sample to detector distances of 0.3 and 6 m, both at a wavelength λ of 6 Å, were used to obtain this range. The neutron spread $\Delta\lambda/\lambda$ for all experiments was 15%. Circular aperture diameters of 40 mm and 12 mm for source and sample, respectively, were used for all measurements. A 2D ($1 \times 1 \text{ m}^2$) ^3He linear position-sensitive detector made up of 192×256 pixels was used to record the data. Detector images were normalized to the incident beam and monitor counts; corrected for pixel sensitivity, detector dark current, and background contributions; and scaled to absolute units.

4.1.5 Measurement of moisture-induced swelling at the bulk level

To measure the swelling of wood at the bulk level, 0.5 mm thick TL sections were conditioned at 75% and 95% RH. Prior to conditioning the samples at 75% RH the samples were conditioned at 25% RH for at least 12 hours. Only the width of all specimens, which corresponded to the tangential direction, was measured using a caliper after the samples were in equilibrium for at least 3 hours. The samples were held flat during measurements to prevent bowing of the samples.

4.2 Results: studies on adhesive infiltrated wood cell walls

4.2.1 Effects of adhesive infiltration on the wood ultrastructure

The SANS 2D scattering patterns obtained from unmodified and dPF-infiltrated wood immersed in a 35:65 D₂O:H₂O solution are shown in Figure 4.3a and b, respectively. As expected, when unmodified wood samples are immersed in the CMP, the contrast between elementary fibrils is lost and the characteristic elementary fibril spacing diffraction peak is no longer visible (Figure 4.3a). Samples that have been modified with dPF have a new source of contrast because dPF has a higher SLD than cellulose as shown in table 4, and thus, still scatter neutrons because of the contrast. The presence of a diffraction peak in the dPF infiltrated sample indicates that adhesives can infiltrate the regions between the elementary fibrils, while preserving most of the order of the elementary fibrils. The reduced 1D scattering profiles for each pattern are shown in Figure 4.3 (c). Because wood is not a two-component system, it is not surprising that the scattering has not entirely removed throughout all the q-range even when the cellulose CMP is used. Scattering from lignin might still contribute because it has a higher SLD than cellulose, Table 4.4. Since using the cellulose CMP removed the scattering beyond $0.01\text{\AA}^{-1}$, this suggests that scattering from lignin is not contributing at this length scale. Whereas, at lower q (below

$0.01\text{\AA}^{-1}$) the presence of a power law suggests that scattering from lignin is contributing at these length scales.

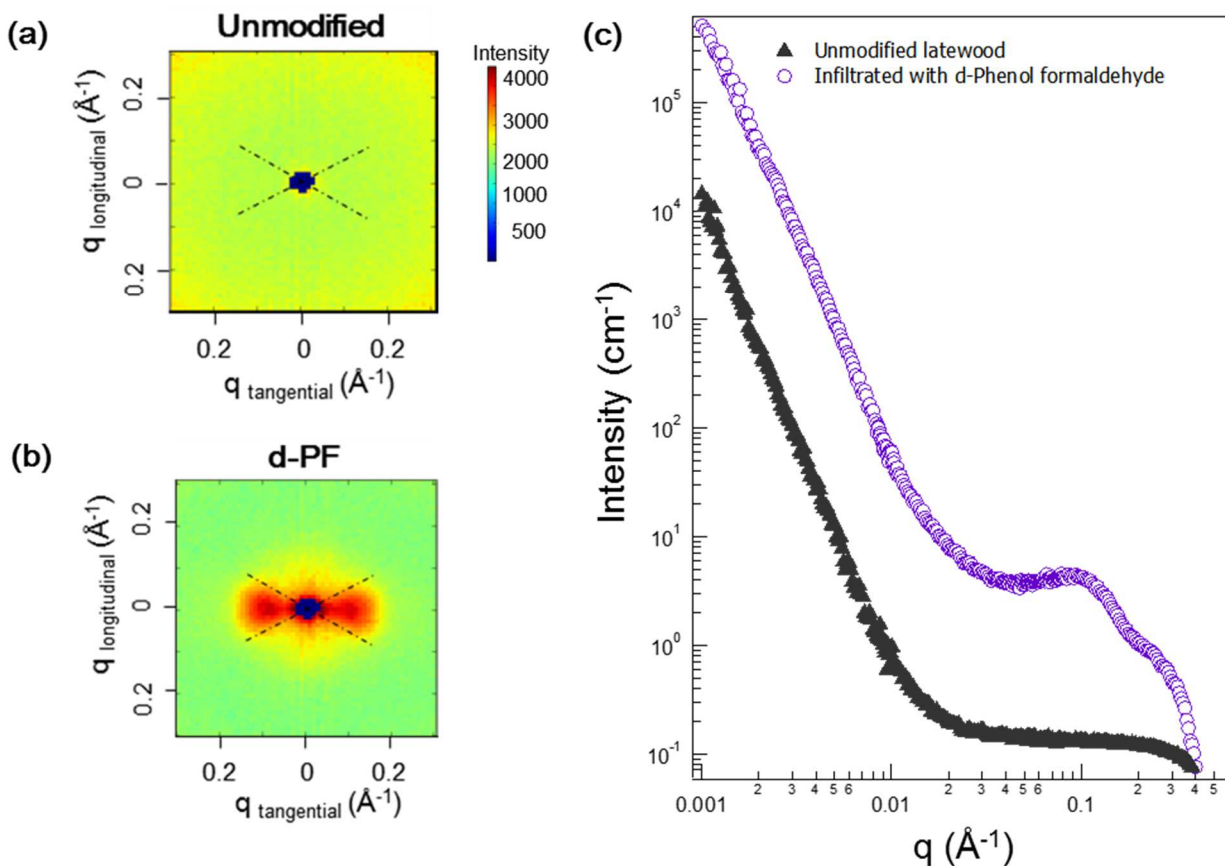


Figure 4.3 Two-dimensional anisotropic scattering patterns from an (a) unmodified latewood sample and (b) a sample infiltrated with deuterated PF, both immersed in a 35:65 $\text{H}_2\text{O}:\text{D}_2\text{O}$ solution. (c) Anisotropically reduced scattering profiles from the equatorial sectors shown in the 2D images using dashed lines.

Table 4.4 Relevant neutron scattering length densities for wood infiltrated with deuterated PF.

Components	Molecular Formula	Density (g/cm ³)	Neutron SLD (10 ¹⁰ cm ⁻²)
Cellulose (crystalline)	C ₆ H ₁₀ O ₅	1.55	1.87
Hemicelluloses	C ₆ H ₁₀ O ₅	1.40	1.62
Lignin	C _{9.92} H _{10.68} O _{3.32}	1.46	2.64
d-PF adhesive	C ₇ D ₇ O ₈	1.5	7.84
35D₂O:65 H₂O	D ₂ O	~1	1.87

4.2.2 Effects on moisture-induced swelling at the elementary fibril level

For unmodified wood under dry conditions, the contrast between the crystalline elementary fibrils and water-accessible matrix where these are embedded is insufficient to resolve the Bragg diffraction peak. Hence, there is no peak in the high q Figure 4.4a. However, samples that have been infiltrated with hydrogenated PF or pMDI exhibit a diffraction peak under dry conditions at this level, Figure 4.4 (c-f). This indicates that in these adhesive-infiltrated samples there is sufficient contrast between the elementary fibrils and the regions between them, meaning, that the adhesives must be infiltrating the regions between the elementary fibrils. The relatively low intensity of the peak can be attributed to having a reduced contrast between the cellulose

elementary fibrils and the infiltrated adhesives, because their scattering length densities (SLDs) are estimated to be smaller than the SLD of D₂O as shown in Table 4.5.

Table 4.5 Relevant neutron scattering length densities for adhesive infiltrated wood.

Components	Molecular Formula	Density (g/cm³)	Neutron SLD (10¹⁰ cm⁻²)
Cellulose (crystalline)	C ₆ H ₁₀ O ₅	1.55	1.87
Hemicelluloses	C ₆ H ₁₀ O ₅	1.40	1.62
Lignin	C _{9.92} H _{10.68} O _{3.32}	1.46	2.64
PF adhesive	C ₇ H ₇ O ₈	1.5	2.75
pMDI adhesive	C ₁₅ H ₁₀ O ₂ N ₂	1.5	3.5
D₂O	D ₂ O	1.11	6.39

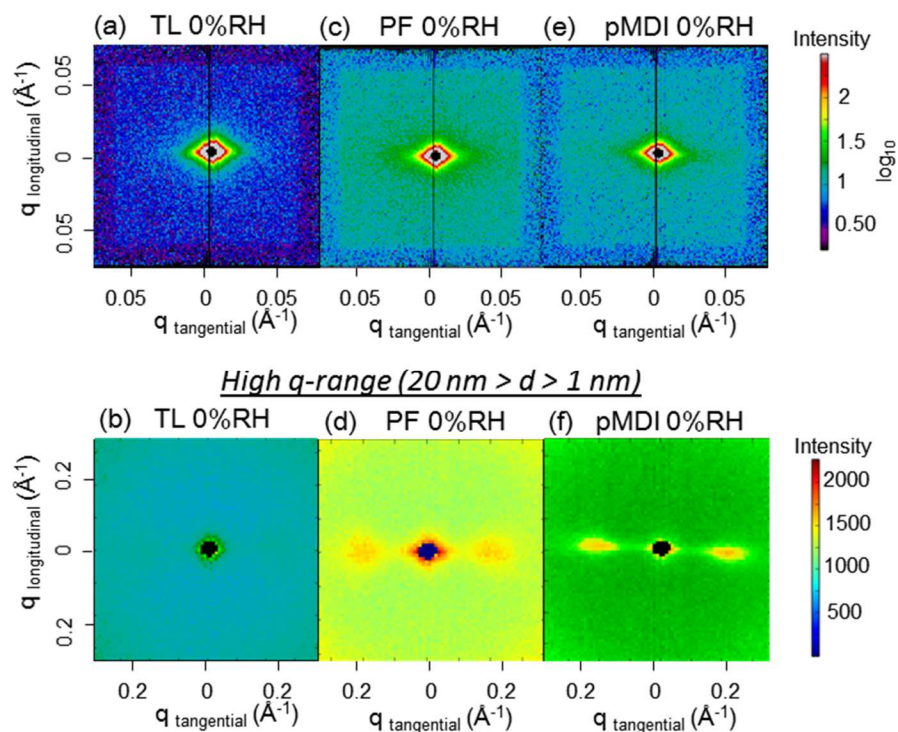


Figure 4.4 Two-dimensional anisotropic scattering patterns over a broad range of scattering vectors obtained from wood samples conditioned at 0% RH. Patterns correspond to measurements from tangential-longitudinal 0.5mm thick wood sections that were (a-b) unmodified (TL), and infiltrated with (c-d) phenol-formaldehyde or (e-f) polymeric methylene diisocyanate (pMDI).

As humidity increases the overall intensity of the scattering pattern of unmodified wood increases due to an increase in contrast as D_2O molecules enter the cell wall, Figure 4.5 (a-b). Interestingly, an increase in intensity is also observed in the samples that have been infiltrated with moisture-durable adhesives. The increased contrast leads to more prominent scattering features in all samples. Phenol-formaldehyde infiltration did not significantly change the scattering features characteristic of unmodified wood, suggesting that this infiltration did not significantly change the cell wall ultrastructure (Figure 4.5 (c-d)). Conversely, pMDI infiltration resulted in higher directional

scattering in the low q range, which suggests that elementary fibrils are more aligned in this sample Figure 4.5 (e-f).

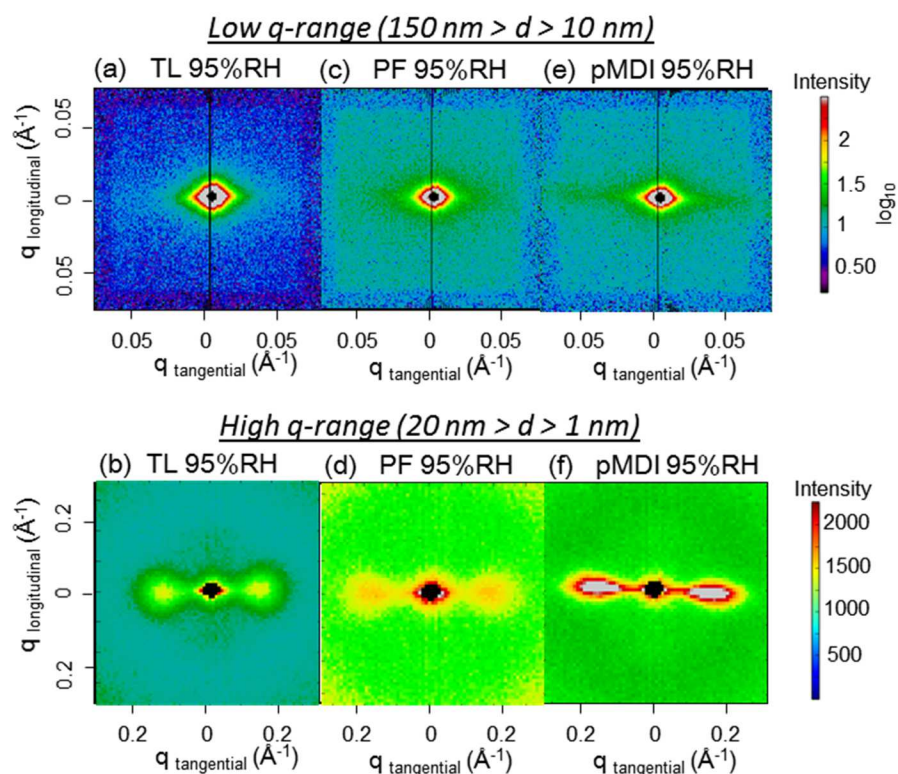


Figure 4.5 Two-dimensional anisotropic scattering patterns obtained from wood samples conditioned at 95% RH. Patterns correspond to measurements from tangential-longitudinal 0.5mm thick wood sections that were (a-b) unmodified (TL), and infiltrated with (c-d) phenol-formaldehyde or (e-f) polymeric methylene diisocyanate (pMDI).

To elucidate the effects of humidity on the elementary fibril spacing the scattering patterns were anisotropically reduced. The 1D scattering profiles obtained from the reduction are shown in Figure 4.6. In these profiles, it is more evident that the elementary fibril spacing peak position shifts to lower q with increasing RH. This shift indicates that as water enters between the

elementary fibril, the center to center distance between the elementary fibrils increases for all samples as a function of humidity, which has been observed previously in unmodified wood.^{20,22}

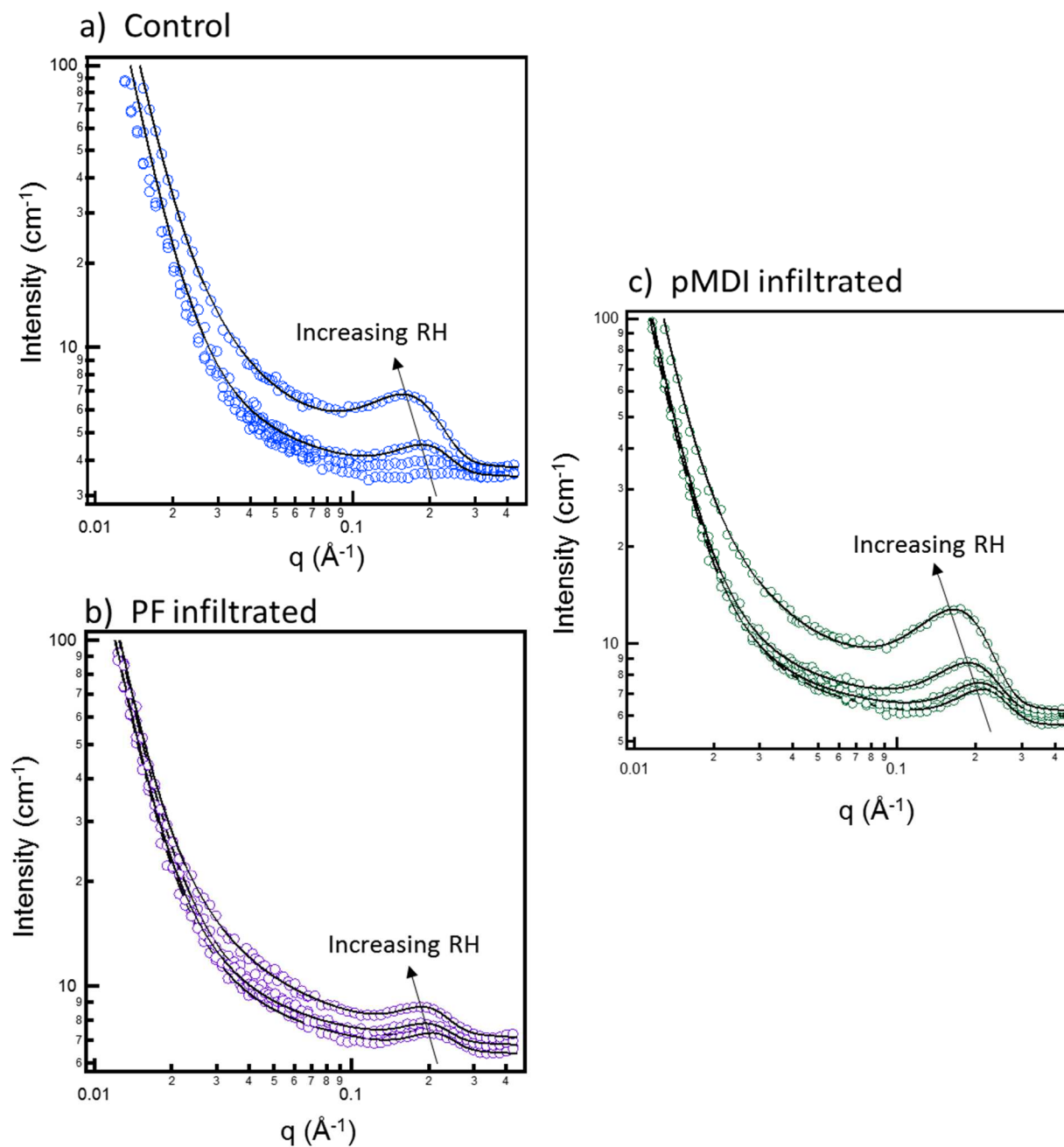


Figure 4.6 Humidity dependence of the scattering profiles from (a) an unmodified latewood sample, and a sample infiltrated with (b) phenol-formaldehyde (PF) or (c) polymeric methylene diisocyanate (pMDI).

To quantify changes in the moisture-induced swelling at the elementary fibril level, the reduced data were fitted to the same empirical model (eq. 3.2) used previously in the SANS studies on the unmodified latewood samples shown in chapter 3.²⁰ The model includes two power laws, a Gaussian peak, and a constant background. Like the previous studies, no humidity dependence was observed in the power law scattering. Furthermore, this trend was not affected by the adhesive infiltration, and no significant differences were observed in the magnitude of the power law slopes. The low q power law ($q^{-4.2 \pm 0.2}$) is thought to be due to smooth interfaces likely due to scattering from the lamellar surfaces of the bundles of elementary fibrils. Whereas, the mid q power law ($q^{-1.2 \pm 0.3}$) is caused by the scattering of the long elementary fibrils.

The position of the diffraction peak due to packing of the elementary fibrils was converted into real space using the relation $d = \frac{2\pi}{q}$ to retrieve the elementary fibril spacing. The humidity dependence of the elementary fibril spacing is shown in Figure 4.7. Interestingly, the elementary fibril spacing was only measurable above 75% RH for the unmodified sample, whereas, adhesive infiltrated samples exhibited a diffraction peak even at dry conditions. The lack of a diffraction peak in the unmodified sample can be attributed to a high order of polydispersity in the alignment of the fibrils with respect to the beam. Subsequent x-ray diffraction experiments confirmed that the latewood ring selected had a high MFA ($>20^\circ$), which resulted in a high degree of misalignment in the irradiated sample volume. In adhesive infiltrated samples, on the other hand,

the adhesive acts as a new source of contrast that contributed to the diffraction peak intensity, particularly at low humidity levels (< 40% RH).

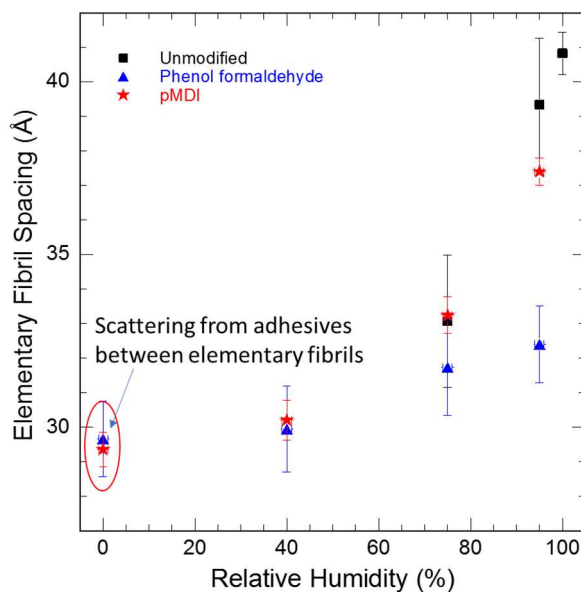


Figure 4.7 The humidity dependence of the elementary fibril spacing found in unmodified latewood, and samples infiltrated with PF and pMDI.

4.2.3 Discussion

The measured elementary fibril spacing in the adhesive infiltrated samples is comparable to the elementary fibril spacing in unmodified samples at low humidity levels. The presence of a diffraction peak in the infiltrated sample at dry conditions when there is no sufficient contrast between the wood polymers indicates that the adhesive can infiltrate the water-accessible regions between the elementary fibrils (Figure 4.8).

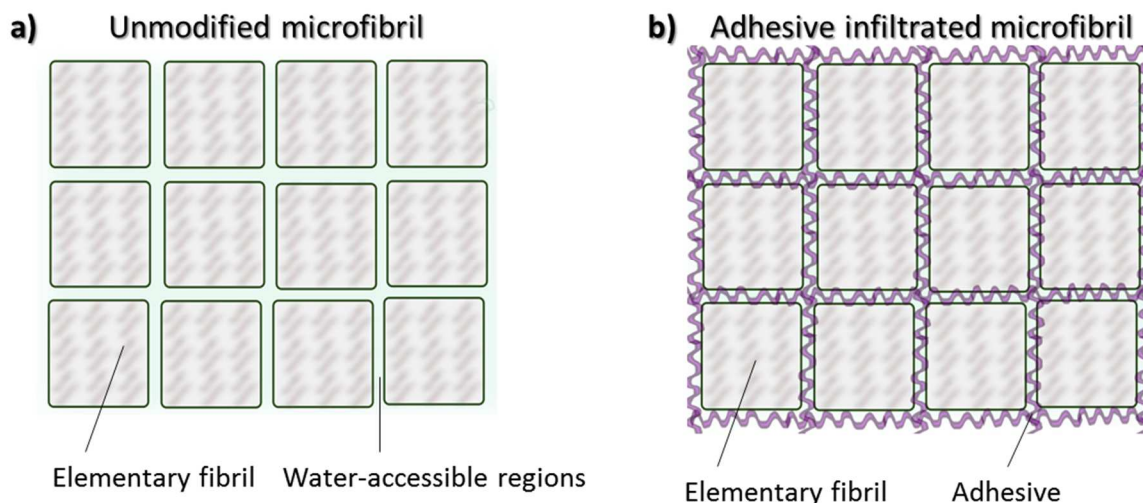


Figure 4.8 Schematic showing the effects on the adhesive infiltration at the elementary fibril level. Here a proposed array of 3x4 elementary fibrils constitutes the microfibril. Transverse view of (a) Unmodified microfibril and (b) an adhesive infiltrated microfibril.

By comparing the effects of adhesive infiltration on the overall dimensional changes from 75 to 95% RH, Table 4.6, it is noticeable that the PF infiltration reduced significantly the swelling at both bulk and elementary fibril levels. Conversely, the pMDI infiltration used in this study did not reduce significantly the moisture-induced swelling. However, neither the amount of adhesive infiltration nor the adhesive molecular weight were controlled; hence, a direct comparison between the two adhesives is beyond the scope of this study. Nevertheless, for these samples there is a direct correlation between the bulk swelling and the swelling at the elementary fibril level. This provides further confirmation that SANS can be used to study wood protection treatments and their ability to address swelling at the elementary fibril level, which is correlated to the swelling at the bulk level.

Table 4.6 Moisture-induced swelling strain from 75 to 95% RH at the bulk and elementary fibril (EF) level.

Sample	Bulk level (mm length scale, %)	EF level (nm length scale, %)
Unmodified	2.28 (0.68)	18.97 (1.44)
PF	0.45 (0.20)	2.11 (0.12)
pMDI	2.12 (0.17)	12.5 (0.24)

These results indicate that moisture-durable adhesives attain at least a portion of their moisture durability by infiltrating the water-accessible regions between the elementary fibrils. This infiltration reduces the sorption capacity of these regions, thus reducing the moisture-induced swelling strain at the elementary fibril level, which consequently reduces the dimensional changes at the bulk level.

While elementary fibril infiltration is evident in this study, the molecular-scale details of the infiltration remain unclear. For instance, from these studies it cannot be determined whether the infiltration of the adhesive led to a mechanical interlocking as the adhesive flowed into the water-accessible spaces or if there was covalent bonding between the wood polymers and the adhesives at the elementary fibril level. Nevertheless, these studies prove that SANS can now be used as a tool to study the effects of concentration (WPG) or molecular weight distribution on the elementary fibril infiltration, as well as the efficacy of the infiltration in preventing the moisture-induced swelling.

4.3 Studies on acetylated wood cell walls

4.3.1 Effects of acetylation on the wood ultrastructure

D-labeled acetylated samples do not exhibit a diffraction peak (Figure 4.9) under dry conditions. The lack of a diffraction peak indicates that even though the deuterated acetyls have a higher SLD than cellulose, Table 4.7, they are not providing contrast between elementary fibrils, meaning acetylation does not occur in the water-accessible regions between the elementary fibrils in such a way that causes fiber diffraction. Despite not being highly correlated, the deuterated acetyls have preferential orientation, which gives rise to the highly aligned equatorial scattering observed. This directional scattering suggests that deuterated acetyls are aligned parallel to the elementary fibrils. Once the data is anisotropically reduced, it is noticeable that the aligned scattering corresponds to power law scattering with a slope equal to -1, which is generally attributed to

scattering from elongated one-dimensional objects like cylinders or rods. This further supports that the deuterated acetyl groups are forming highly elongated regions that are aligned parallel to the cellulose elementary fibrils. At lower q both samples have power law scattering with a slope of -4, which is indicative of scattering from smooth surfaces or interfaces. However, the cross-over between the power laws is shifted to a lower q in the acetylated sample, which might be indicative that the smaller dimension of the object, whose surface gives rise to the low q power law, is increasing.

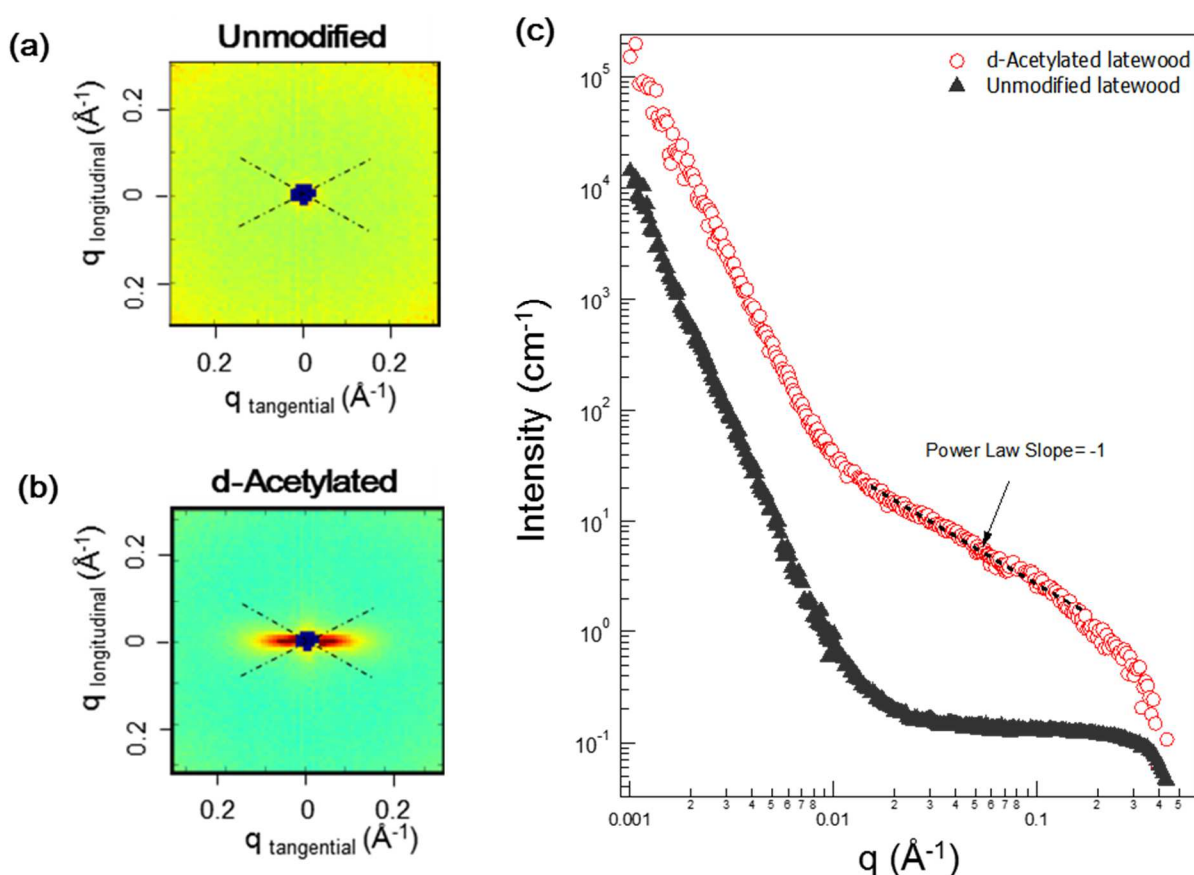


Figure 4.9 Two-dimensional anisotropic scattering patterns from an (a) unmodified latewood sample and (b) a sample modified with deuterated acetyls, both immersed in a 35:65 $\text{H}_2\text{O}:\text{D}_2\text{O}$ solution. (c) Anisotropically reduced scattering profiles from the sectors shown in the 2D images using dashed lines.

Table 4.7 Relevant scattering length densities in wood cell walls acetylated with deuterium labeled acetyls.

Components		Molecular Formula	Density (g/cm ³)	Neutron SLD (10 ¹⁰ cm ⁻²)
Cellulose (crystalline)		C ₆ H ₁₀ O ₅	1.55	1.87
Hemicelluloses	Hexose	C ₁₂ H ₁₈ O ₉	1.5	1.50
	d-Acetylated hexose	C ₁₂ H ₉ D ₉ O ₉	1.5	3.01
Lignin	Conyferyl alcohol	C ₁₂ H ₁₈ O ₉	1.5	1.96
	d-Acetylated conyferyl alcohol	C ₁₄ H ₁₀ D ₆ O ₇	1.5	4.07

4.3.2 Effects on moisture-induced swelling at the elementary fibril level

The two-dimensional scattering patterns obtained from hydrogenated samples at a low humidity level, namely, 25% RH are shown in Figure 4.10. At lower q , (Figure 4.10 a, c, e and g), where scattering from the lamellar structures made of bundles of cellulose elementary fibrils is expected to be the major contributor, the scattering patterns are characterized by diamond-shaped scattering that becomes more pronounced as acetylation increases. This suggests that at this length scale the contrast increased with increased acetylation. This increase can be attributed to having an increase in the amount of acetylated polymers, which have a higher SLD, Table 4.8.

Conversely, at high q , the diffraction peak due to the elementary fibril packing is only visible in unmodified samples and no further differences were observed as a function of acetylation.

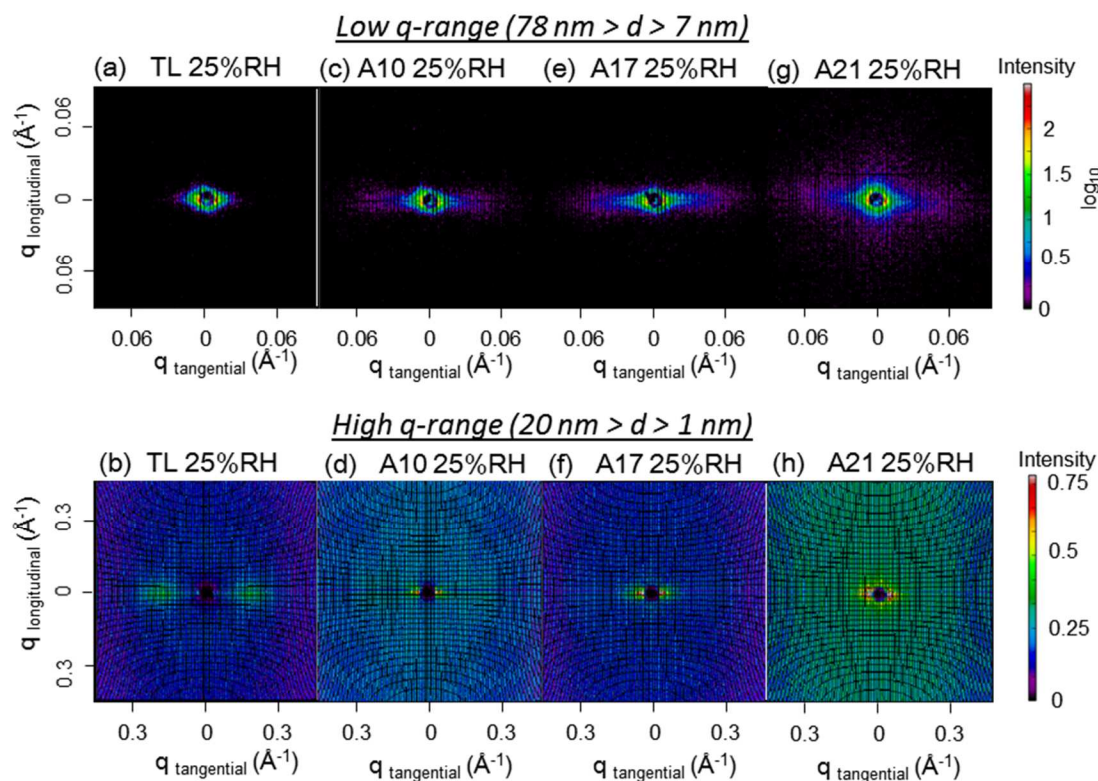


Figure 4.10 Two-dimensional anisotropic scattering patterns obtained from samples conditioned at 25% RH. Patterns correspond to measurements from tangential-longitudinal 0.5 mm thick wood sections that were (a-b) unmodified (TL), and modified with (c-d) 10WPG (A10), (e-f) 17WPG (A17) and (g-h) 21WPG (A21) acetylation.

Table 4.8 Relevant scattering length densities in acetylated wood cell walls.

Components		Molecular Formula	Density (g/cm ³)	Neutron SLD (10 ¹⁰ cm ⁻²)
Cellulose (crystalline)		C ₆ H ₁₀ O ₅	1.55	1.87
Hemicelluloses	Hexose	C ₁₂ H ₁₈ O ₉	1.5	1.50
	Acetylated hexose	C ₁₂ H ₁₈ O ₉	1.5	1.91
Lignin	Conyferyl alcohol	C ₁₂ H ₁₈ O ₉	1.5	1.96
	Acetylated conyferyl alcohol	C ₁₄ H ₁₆ O ₇	1.5	2.25

The two-dimensional scattering patterns obtained from the same samples at a higher humidity level, namely 95% RH, are shown in Figure 4.11. Here it is shown that when humidity increases the scattering features become more prominent throughout the entire q range. At low q , the diamond-shape scattering is still present in all samples. At this humidity, the increased acetylation also led to a higher degree of alignment in the low q equatorial scattering. At high q , the intensity of the diffraction peak decreases with increasing acetylation. This suggests that the contrast at the elementary fibril level decreases with increasing acetylation perhaps because the regions between the elementary fibrils become less water-accessible.

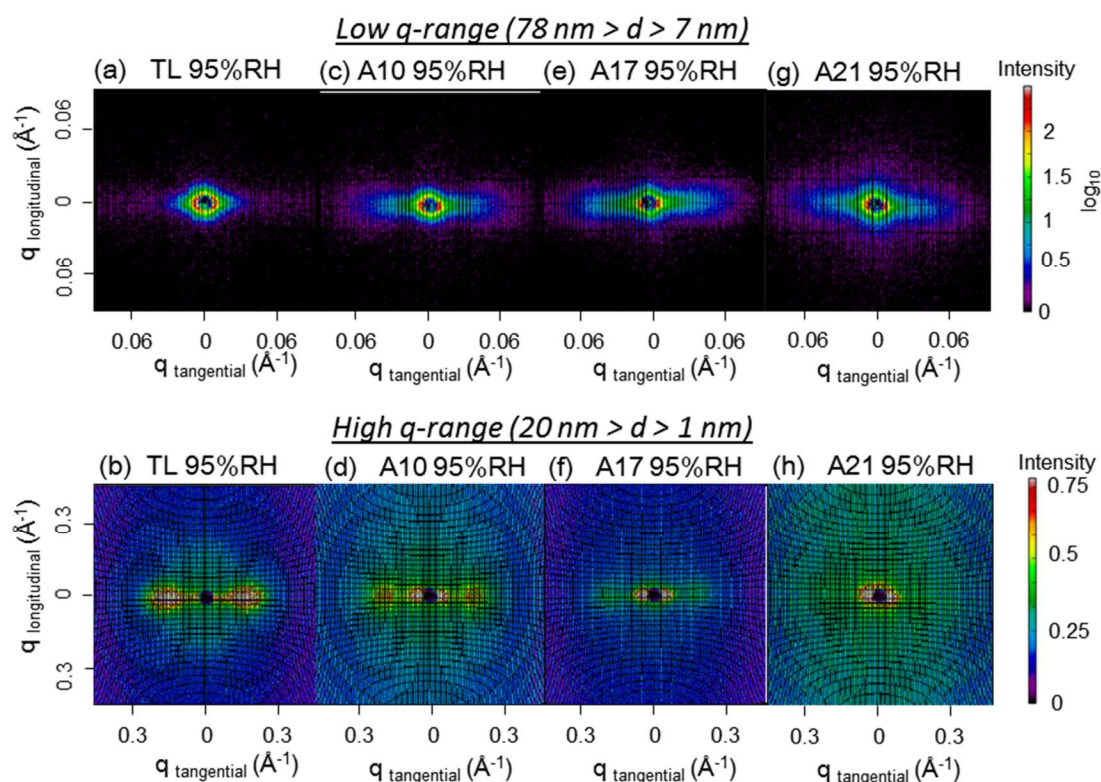


Figure 4.11 Two-dimensional anisotropic scattering patterns obtained from samples conditioned at 95% RH. Patterns correspond to measurements from tangential-longitudinal 0.5mm thick wood sections that were (a-b) unmodified (TL), and modified with (c-d) 10WPG (A10), (e-f) 17WPG (A17) and (g-h) 21WPG (A21) acetylation.

The effects of acetylation on the scattering pattern, particularly at the elementary fibril level, are more evident in the anisotropically reduced one-dimensional scattering data (Figure 4.12). Here, it is noticeable that as acetylation increases the diffraction peak decreases until it is no longer visible in samples with 21% WPG acetylation. While the scattering that gives rise to the Gaussian peak decreased, the mid-q power law scattering increased due to acetylation. This suggests that while the contrast between elementary fibrils decreases, the contrast at higher length scales increases. This trend could be attributed to the amorphous regions between the elementary fibrils becoming less accessible to the water molecules entering the cell wall.

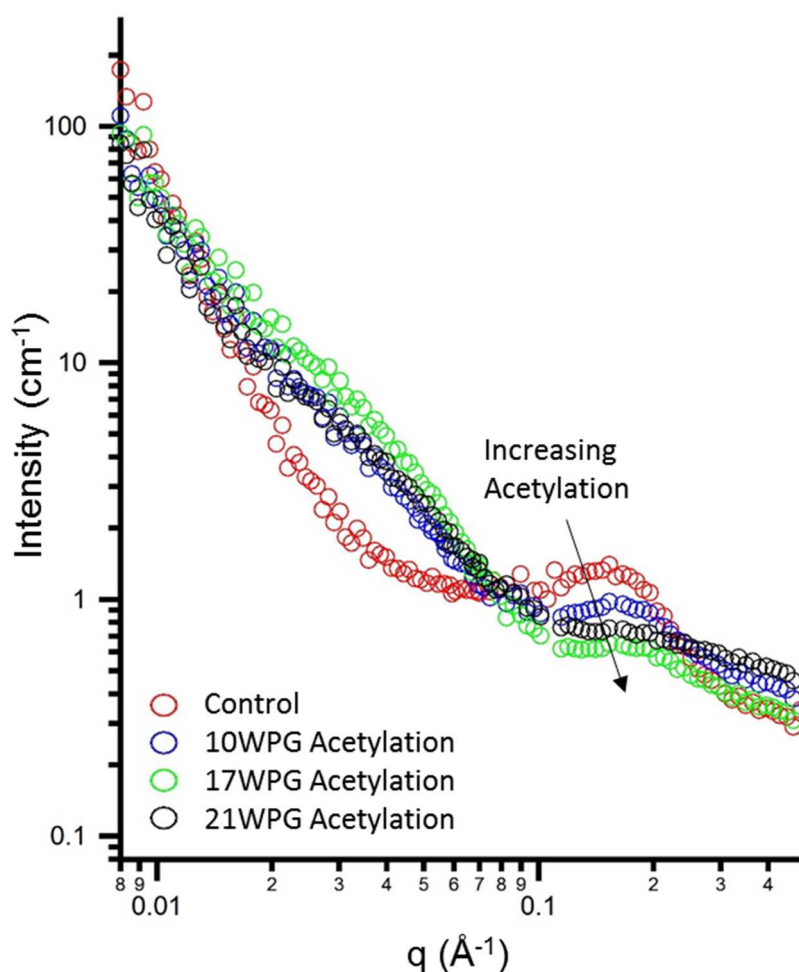


Figure 4.12 Effects of acetylation level on the scattering profile. All curves correspond to samples conditioned at 95% RH.

Overall, the scattering intensity increased with increasing humidity for all samples as shown in Figure 4.13. The relative humidity at which the diffraction peak became visible was dependent on the degree of acetylation, and as acetylation increased higher levels of humidity were necessary for the peak to become visible. The diffraction peak shifted towards lower q with increasing humidity for all samples with acetylation levels below 21% WPG. This trend has also been observed in unmodified wood.^{20,22}

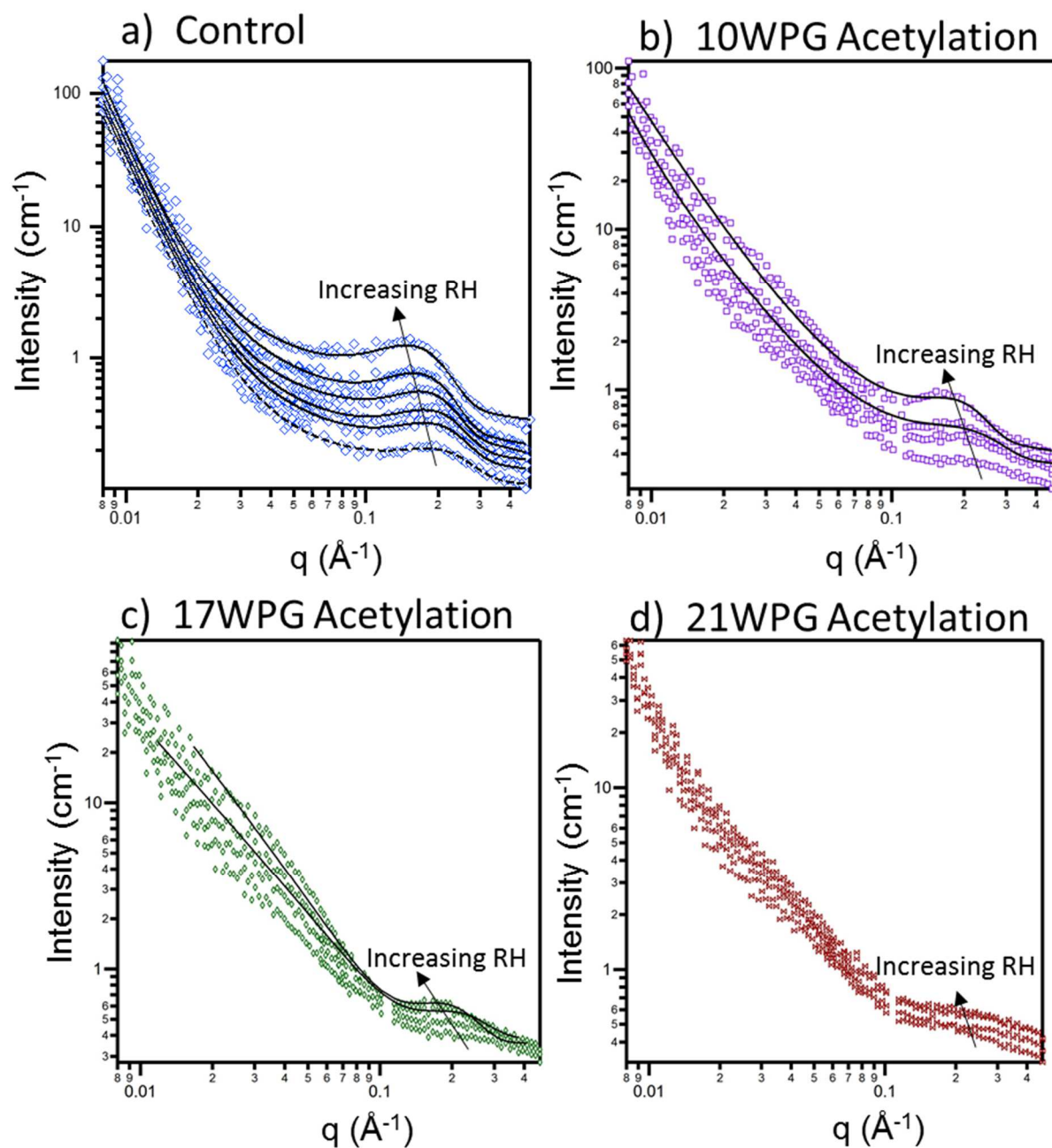


Figure 4.13 Effects of humidity on the samples with different levels of acetylation, ranging from (a) no acetylation to (b) low (10WPG), (c) medium (17WPG) and (d) high (21WPG) acetylation.

The effect of acetylation on the moisture-induced swelling was quantified using the same model described in chapter 3, equation 3.2, which consists of two power laws, a Gaussian peak and a constant background. Here, since no trend was observed in the mid q power law slope it was

held fixed at -1 to decrease the number of independent variables allowed to vary during the fitting. The lower q power law slope showed no humidity dependence, and this trend was observed regardless of the degree of acetylation. However, it was observed that acetylation decreased the low q power law slope from -4 to approximately -2, Table 4.9. Scattering from smooth surfaces of large objects, whose size cannot be fully resolved, leads to power law scattering with a slope of -4, whereas, the slope of -2 is generally associated to scattering of planar objects such as laminae or lamellae.

Table 4.9 Effects of acetylation on the low q power law slope.

ACETYLATION LEVEL (WPG)	LOW Q POWER LAW SLOPE (AVERAGE)	UNCERTAINTY
0	4.08	0.43
10	2.50	0.42
17	1.82	0.14
21	1.85	0.11

The changes in the elementary fibril spacing are shown in Figure 4.14 as a function of humidity as well as moisture content. By tracking the elementary fibril spacing as a function of humidity it is evident that acetylation decreased the overall elementary fibril spacing measured at all humidity levels. This might be attributed to acetylated samples being less hygroscopic, which results in samples that have lower moisture contents.²³ For instance, at 75% RH the unmodified wood sample has a moisture content of 12%, while a sample with 10WPG acetylation has a 7.5% MC and the sample with 21% WPG only has 5% MC. Hence, as acetylation increases the amount of water between the elementary fibrils should also decrease. By tracking the MC dependence of the elementary fibril spacing, Figure 4.14 (b), it can be observed that acetylation does not reduce significantly the elementary fibril spacing as a function of MC. Moreover, the elementary fibril spacing is directly proportional to the moisture content of the sample.

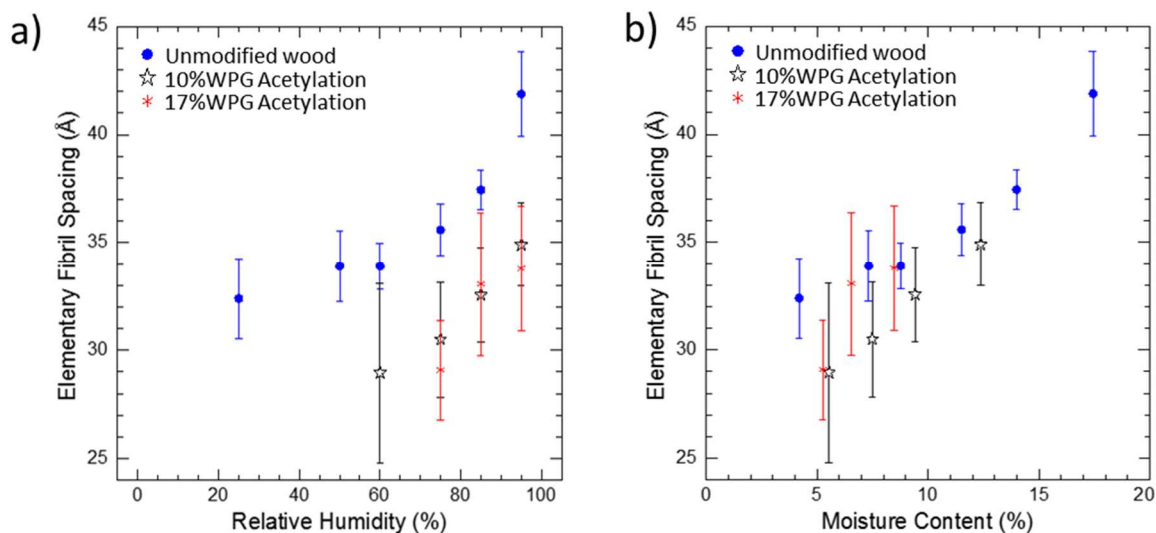


Figure 4.14 Elementary fibril spacing dependence on a) relative humidity of the sample environment and b) moisture content of the sample.

4.3.3 Discussion

The decrease in the intensity of the diffraction peak due to elementary fibril packing as a function of acetylation suggests that this treatment prevents water from entering the water-accessible between the elementary fibrils by acetylating the surrounding regions, Figure 4.15. Acetic anhydride is most reactive with lignin and hemicellulose, which could explain why acetylation is not targeting the regions between the elementary fibrils.¹⁶ Only low levels of acetylation (~8.5% WPG) are needed to acetylate most of the lignin, whereas the hemicelluloses are fully acetylated only at high levels of acetylation (~18 – 21 WPG).¹⁶ Hence, as the acetylation level increases, the total amount of acetylated regions outside the microfibril increase, which further prevents water from entering the water-accessible regions between the elementary fibrils, thus reducing the diffraction peak intensity until the peak is no longer observable.

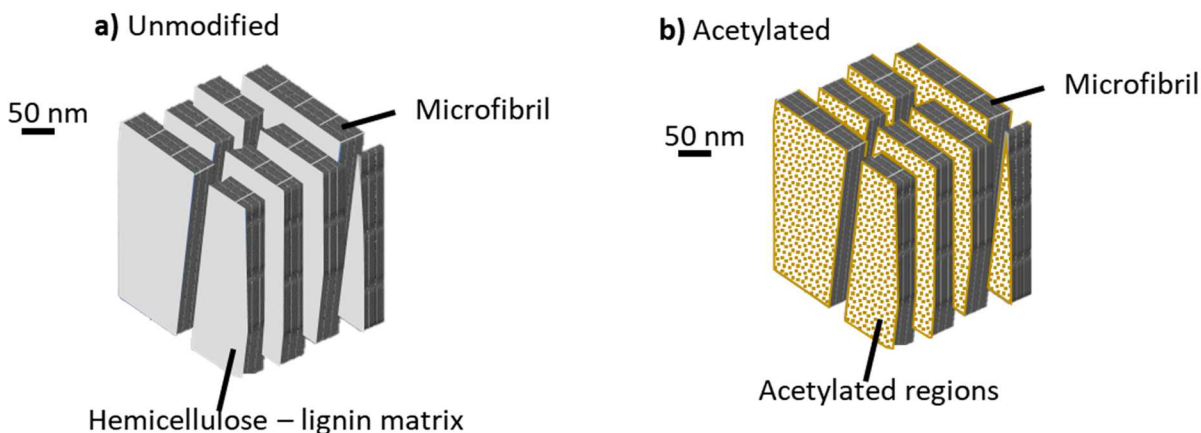


Figure 4.15 Schematic showing the effects of acetylation on the S2 cell wall nanostructure conditioned at 0% RH. (a) Unmodified lamellar structure made of cellulose microfibrils. (b) Lamellar structure showing regions outside the microfibrils being acetylated.

A direct comparison on the effects of acetylation on the moisture-induced swelling across length scales, Table 4.10, shows that from 75% to 95% RH acetylation only reduced significantly the dimensional changes at the bulk level. At the elementary fibril level, however, the measured swelling strain was not significantly reduced beyond the uncertainty of the calculation. The high uncertainty in the strain at the elementary fibril level might be attributed to the decreasing intensity of the diffraction peak with acetylation, which effectively diminished our ability to resolve and fit the peak.

Table 4.10 Moisture-induced swelling strain from 75 - 95% RH at the bulk and elementary fibril (EF) levels. Mean values reported, and errors are shown in parenthesis. Errors at the bulk level were obtained as the standard deviation of measurements taken at the three different locations in the sample, while errors at the EF level were obtained via an uncertainty analysis of the fit.

Sample	Bulk level (mm length scale, %)	EF level (nm length scale, %)
Unmodified	2.1 (0.3)	17.7 (6.5)
10WPG Acetylation	0.7 (0.3)	14.4 (10.9)
17WPG Acetylation	0.7 (0.2)	16.1 (12.8)
21WPG Acetylation	0.4 (0.1)	--

4.4 Conclusions

Chemically modified cell walls can still scatter anisotropically regardless of the type of chemical modification. Contrast variation experiments on D-labeled adhesive infiltrated samples showed adhesives can infiltrate the water-accessible regions between the elementary fibrils. Whereas similar experiments on samples acetylated with deuterium labeled acetyls showed that acetyls cannot infiltrate these regions.

Studies on the moisture-induced swelling of chemically modified cell walls showed that there are two ways to prevent swelling at the bulk level: (1) modify the surroundings of the elementary fibrils to prevent water from entering the regions between the elementary fibrils or (2) infiltrating the water-accessible regions between the elementary fibrils, thus reducing the volume available for water molecules to occupy.

Adhesive infiltration effectively reduced the swelling at the elementary fibril whereas acetylation reduced the amount of water entering the cell wall, thus decreasing the range where the elementary fibril spacing could be measured.

4.5 References

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Chapter 5. Dynamics of water absorbed in unmodified wood cell walls

When wood absorbs water, it leads to structural changes in the cell wall as well as changes in the dynamics of the hydrated polymers and the absorbed water. Despite the importance of understanding the moisture-induced dynamical changes, studies in this area have been limited to measuring the moisture-induced mobility of cellulose¹ and estimating the diffusion coefficient of water in the cell wall based on water absorption² and NMR spin-spin relaxation studies.³ This latter topic is source of debate because previous studies have only been capable of measuring dynamics of free water, while bound water dynamics have been estimated.^{2,3} Recent molecular dynamics simulations have attempted to address this gap by simulating the sorption behavior of hydrophilic wood polymers.⁴ However, no experimental evidence has yet supported the “stop and go” diffusive motion of bound water described by the simulations.⁵ QENS is uniquely suited to provide the experimental evidence necessary to validate these models, because it can probe the same length and time scales accessible in the MD simulations. However, despite its suitability for lignocellulosic research, QENS has only been used to study the temperature dependent dynamics of cellulose.⁶ While studies with humidity control are scarce, QENS is routinely used to study the dynamics of confined water in various systems including cement^{7,8}, micelles⁹, glass¹⁰ and uranyl fluoride¹¹.

In this chapter, the use of QENS to study the dynamical state of water absorbed in the cell wall will be discussed. The data analysis will be presented, and the measured diffusive motions will be compared to the values reported in the simulation literature. Though these studies have only focused on unmodified latewood samples, the methods outlined in this chapter will be useful in studying how the moisture-induced dynamics in the cell wall are affected by chemical and thermal modifications meant to impart moisture-durability.

5.1 Methods

5.1.1 Sample preparation

Tangential-longitudinal latewood sections (0.3 mm thick) of kiln dried loblolly pine (*Pinus taeda*) were prepared for QENS from a single growth ring using a sled microtome fit with a disposable microtome blade. Figure 5.1(a) shows how several sections were placed next to each other to fill the area illuminated by the beam (3 X 3 cm²).

5.1.2 Humidity chamber

A humidity chamber, custom built from aluminum (Figure 5.1b) was used to test the moisture-induced dynamics of the sample at various humidity conditions ranging from 3.5% to 98% RH. Latewood sections were secured inside the chamber with fasteners. (Figure 5.1 (a)) The humidity inside the chamber was controlled using an L&C Science and Technology (Hialeah, FL, USA) RH-200 relative humidity generator, with a nitrogen (99.99%) cylinder and an external reservoir filled with deionized water. The RH-200's dew point analyzer method was used to control the humidity, meanwhile the temperature and humidity were measured *in situ* using a SHT21x sensor from Sensirion [Staefa ZH, Switzerland].

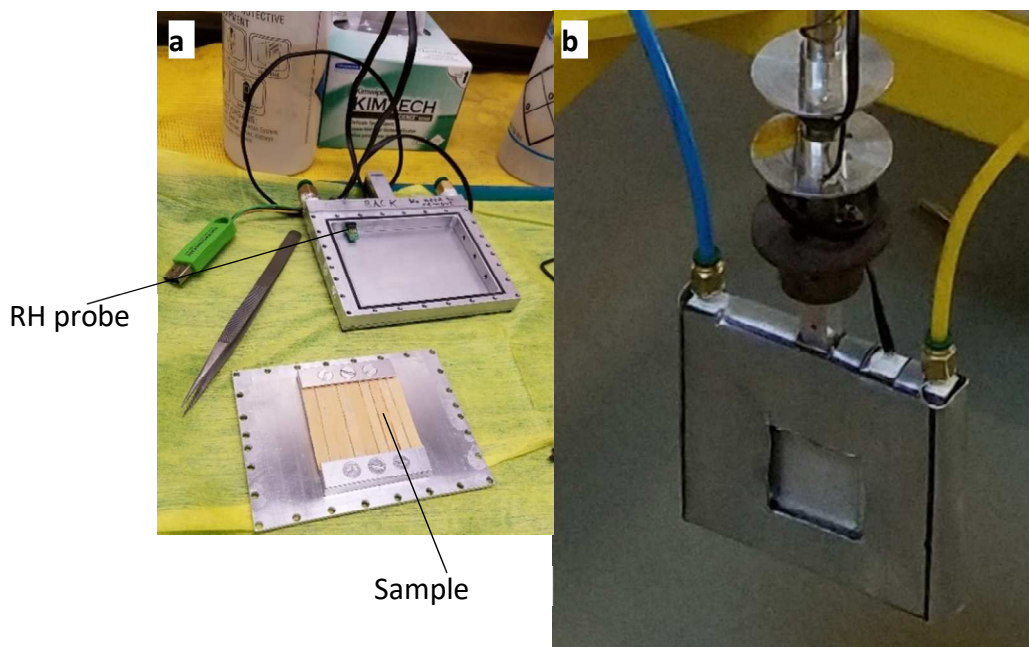


Figure 5.1 Humidity chamber used at BASIS. (a) Open humidity cell showing the sample illuminated area (b) closed humidity chamber with Cadmium mask. The humidity chamber was built at the FPL machine shop.

5.1.3 Quasielastic neutron scattering (QENS) experiments

Humidity controlled quasielastic neutron scattering experiments were performed at the Oak Ridge National Laboratory Spallation Neutron Source on the backscattering BASIS spectrometer with an energy resolution of $1.75\mu\text{eV}$ ($\sim 400\text{ps}$) and a dynamic range of $\pm 120\mu\text{eV}$. Raw spectra were normalized to a vanadium standard, and binned in 9 momentum transfer (q) groups with a binning step size of $0.25\text{ \AA}^{-1}$ from $0.2\text{ \AA}^{-1}$ to $2\text{ \AA}^{-1}$. Data were collected in scans of 15-minute intervals to track the evolution of the dynamics of the sample as it absorbed water molecules. Samples were conditioned for one hour at each RH step, then once the sample dynamics observable within the dynamic range of the instrument were in equilibrium, as determined by comparing consecutive 15-minute intervals, data were collected for four hours to improve the count statistics. Although, coherent diffraction from cellulose was expected to contribute to the signal, particularly around $q \approx 1.1\text{ \AA}^{-1}$, and consequently, shorten the q range that could be analyzed, cellulose coherent

diffraction was not significant. At low angles ($q < 0.4 \text{ \AA}^{-1}$) small angle scattering from the cellulose elementary fibrils¹² contributed to the signal, thus the QENS broadening was only analyzed from $0.45 \text{ \AA}^{-1}$ to $2 \text{ \AA}^{-1}$

5.2 Results: QENS of unmodified wood

The mobility of atoms with large incoherent cross-sections, which for wood that means all the hydrogen atoms in the sample, gives rise to the QENS signal. Hydrogens that are moving much slower than the instrument resolution contribute to the elastic intensity, and hydrogens moving much faster than the available dynamic range will contribute to the background. Selected QENS spectra ($q = 0.95 \text{ \AA}^{-1}$) collected on BASIS as a function of relative humidity are shown in Figure 5.2(a). The similarities between the datasets at low relative humidity show that below 20% RH there was no observable QENS broadening. In Figure 5.2(b) the lack of observable broadening at low humidity is shown by comparing the low humidity spectrum with the instrument resolution, which was measured using a Vanadium standard.

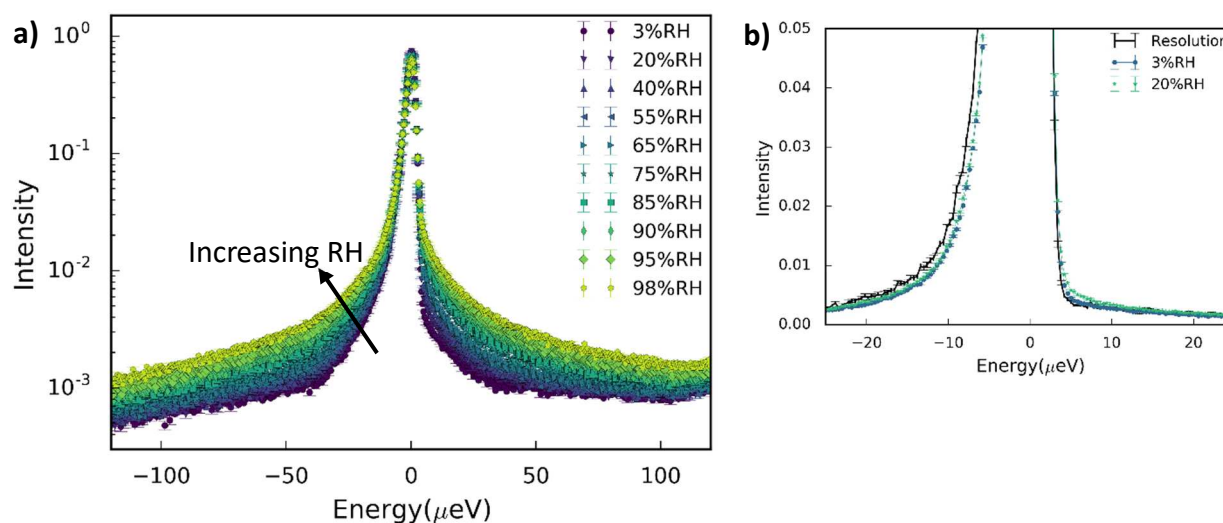


Figure 5.2 (a) Quasielastic spectra as a function of RH (b) linear plot of the spectra of RH steps below 40% RH compared to the resolution of the instrument.

To better understand the origins of the QENS signal, we tracked the moisture-induced changes in the total intensity by integrating the area under the curve and averaging over the measured q -range. The total intensity increased as a function of humidity (Figure 5.3).

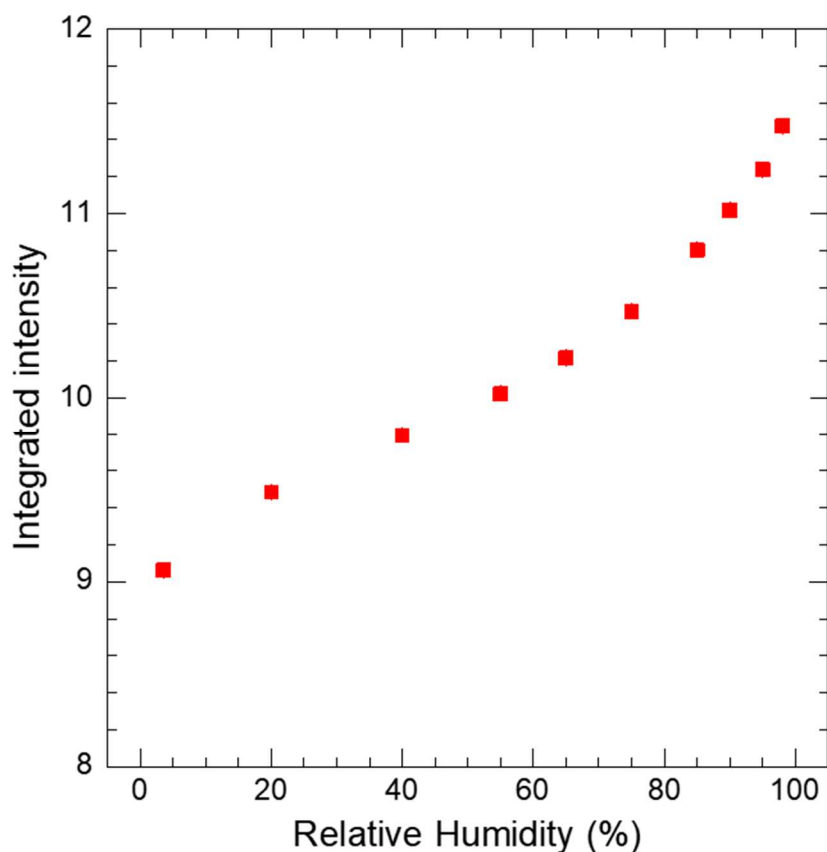


Figure 5.3 Total integrated intensity over all q -range as a function of humidity.

The increase in intensity is expected to be proportional to the change in the number of hydrogenated molecules in the sample. The moisture enhanced mobility of the polymers would only be expected to contribute to an increase in the background, and because the background is included in the total integrated intensity the mobility of the polymers could contribute to the total increase in intensity. However, since only a small fraction of them would become mobile, their contribution to the total intensity is not expected to be significant.⁶ The contribution of the

hydrogens from the dry wood was removed by subtracting the total intensity measured at the lowest humidity (3% RH). Then, the intensity increase due to moisture uptake was calculated as:

$$\Delta I \% = \left(\frac{I(RH) - I(RH = 3\%RH)}{I(RH = 3\%RH)} \right) * 100 \quad \text{eq. 5.1}$$

Figure 5.4 shows that the change in intensity ΔI agrees with the moisture content of Loblolly pine as calculated from the absorption isotherm¹³. This indicates that the increases in the total intensity occurs due to the moisture uptake in the sample.

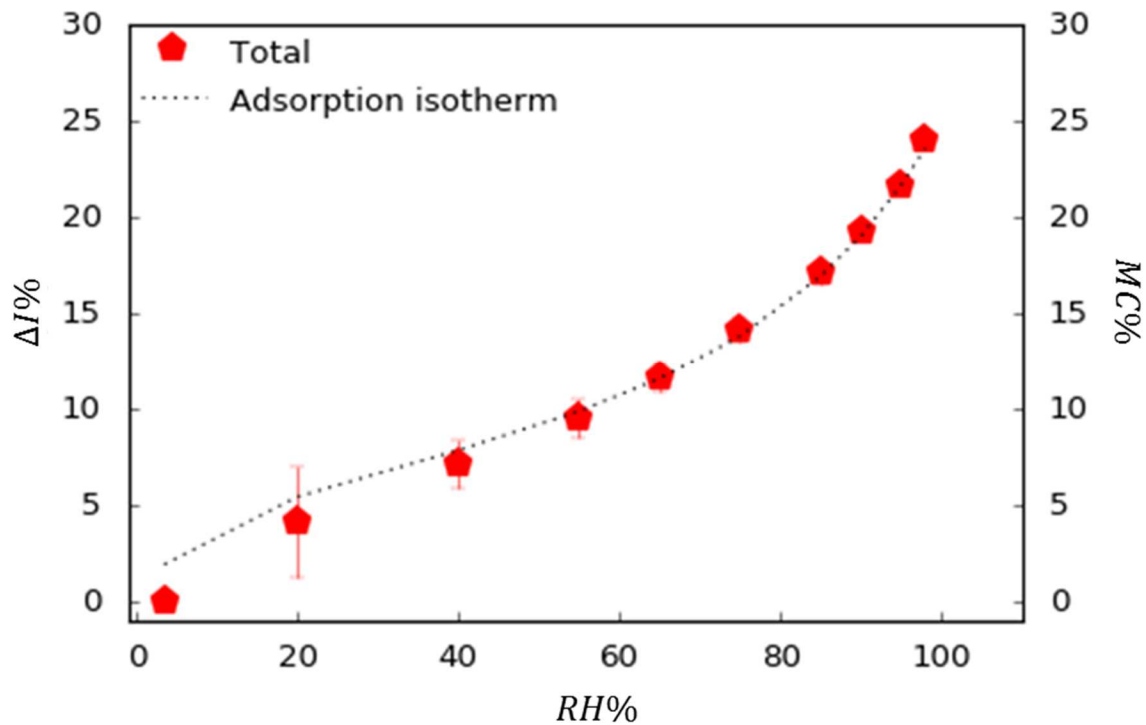


Figure 5.4 Change in total intensity due to addition of water molecules.

Furthermore, by comparing the total integrated intensity to the elastic and quasielastic contributions it can be observed that the elastic region is not contributing to the overall increase, Figure 5.5. Hence, since the background contribution is negligible, the total intensity increase

can be attributed to an increase in the QENS signal. Thus, the moisture increase in the sample is giving rise to the QENS broadening.

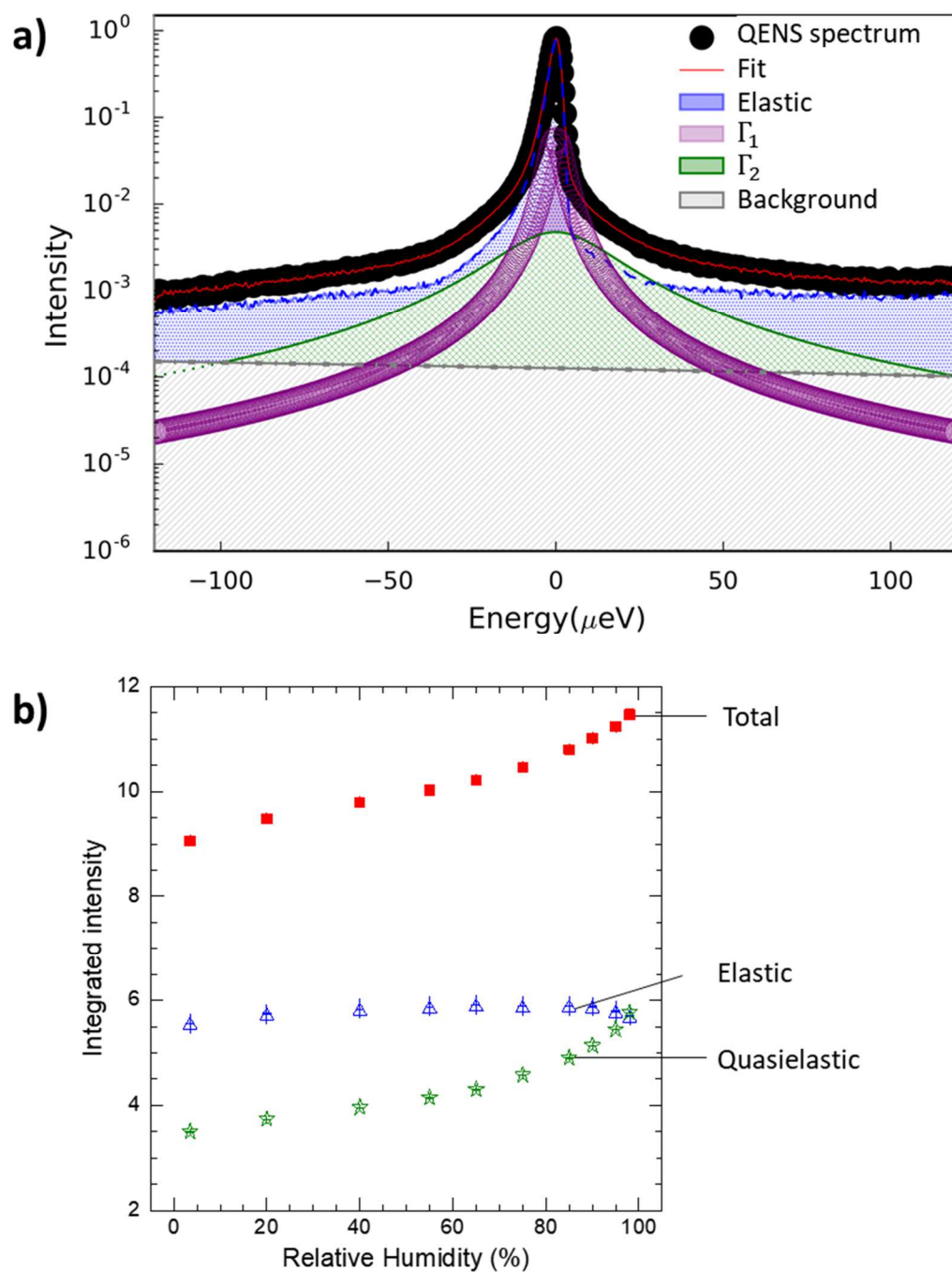


Figure 5.5 a) QENS spectra showing the integrated areas of the elastic quasielastic contributions (,). Data were obtained from latewood sample conditioned at 85% RH, and binned from 0.65 to 0.85 $\text{\AA}^{-1}$. (b) Comparison between the total intensity and its elastic and quasielastic contributions as function of RH.

5.2.1 Analysis of the elastic intensity

By integrating the QENS spectra within the energy resolution of BASIS we can obtain the elastic responses as a function of relative humidity, Figure 5.6. The decrease in the elastic intensity with increasing humidity at high q indicates that the dynamics of the system entered the energy resolution available at the instrument. This approach has been used previously to study phase transitions and/or dynamic cross-overs in proteins¹⁴ as well as transitions in water confined in carbon nanotubes as a function of temperature.¹⁵

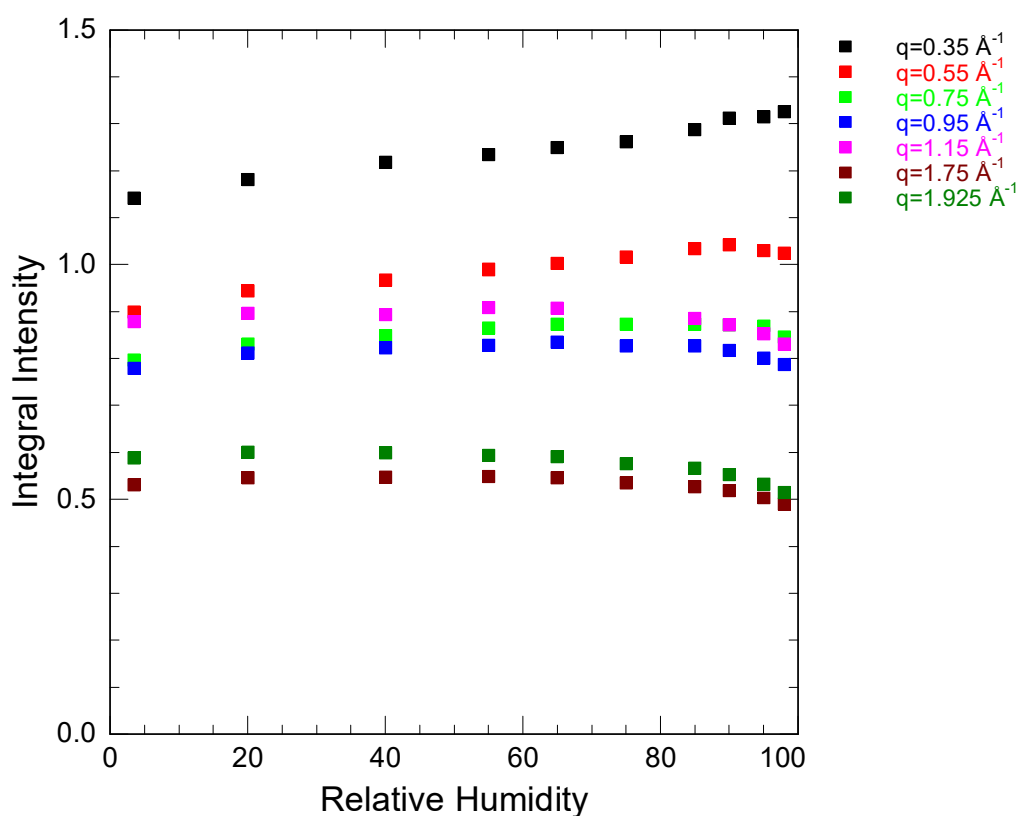


Figure 5.6 Integrated elastic intensity curves for each q value as a function of humidity.

By normalizing the elastic incoherent scattering intensity with respect to the lowest relative humidity measured, we can obtain the static structure factor, and by using a Gaussian

approximation we can retrieve the mean square displacement (MSD) of the hydrogen nuclei in the sample:

$$\frac{\int_{-\Delta\omega}^{\Delta\omega} S_{el}(q, \Delta\omega, RH)}{S(q, RH \sim 0\%)} = \exp \left[-\frac{q^2 MSD}{6} (q, \Delta\omega, RH) \right] \quad \text{eq. 5.31}$$

Here $\Delta\omega$ is the energy resolution of the instrument, S_{el} is the elastic intensity and S is the static structure factor. The humidity dependence of the obtained static structure factors is shown in Figure 5.7. Initially, S increases with humidity, which can be attributed to an increase in the number of hydrogens in the sample that are not moving within the energy resolution of the instrument. Interestingly, this trend continues as humidity increases for low q values ($q < 0.55 \text{ \AA}^{-1}$). At these low q values, the small angle scattering from the elementary fibrils becomes more dominant and its contributions are likely causing the increase observed. Additionally, confined water that is immobile within the instrument resolution can also contribute to this increase. For higher q values ($q > 0.55 \text{ \AA}^{-1}$), S decreases with increasing humidity above 55% RH. This occurs because the total number of mobile hydrogens in the sample is increasing.

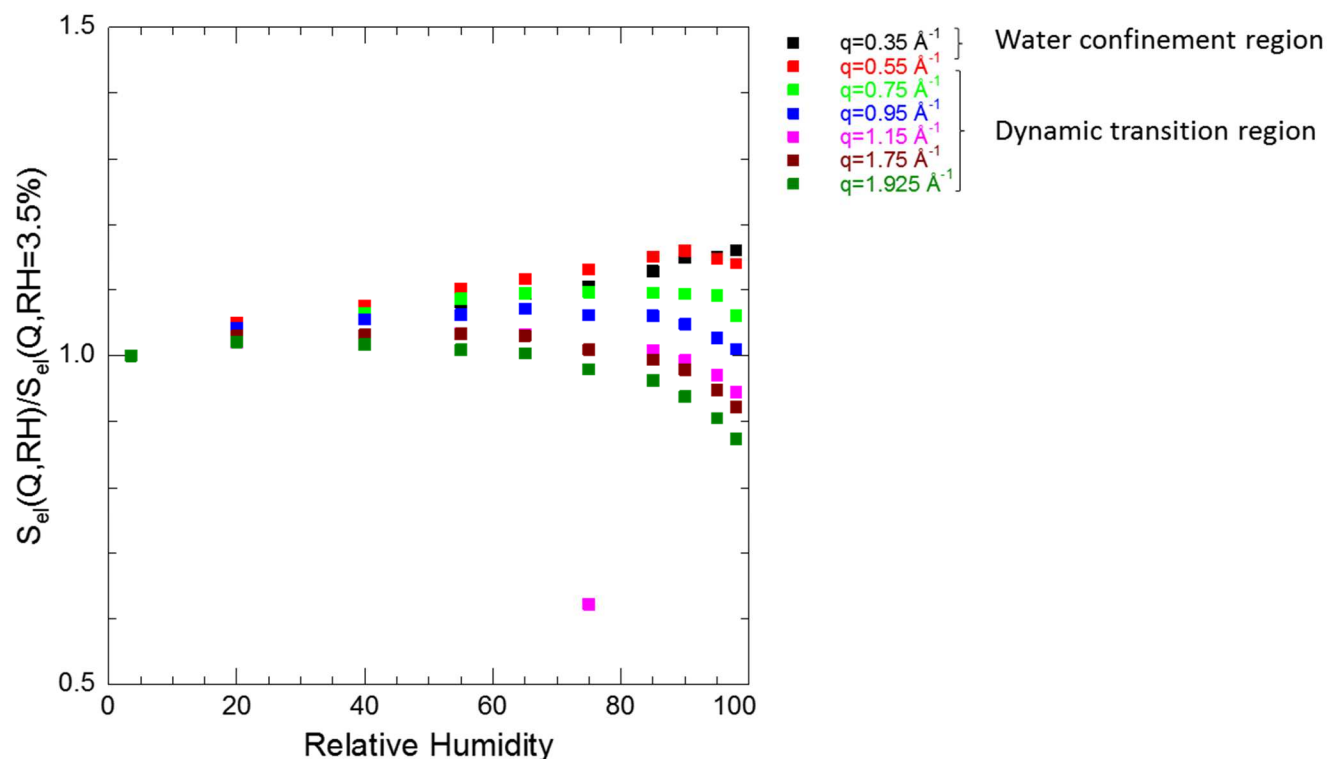


Figure 5.7 Static structure factor as a function of humidity.

The increase in the moisture-induced oscillations of all the hydrogens in the sample is shown in Figure 5.8. At low humidity, the MSD obtained is similar to the values reported for dry wood polymers ($0.1 - 0.2 \text{ \AA}^2$),^{16,17} but as humidity increases the MSD is much larger than the values reported for hydrated cellulose⁶ and lignin¹⁸ (0.2 and $0.5 \text{ \AA}^2$, respectively), suggesting that the relative contribution of the water molecules mobility is much higher than the contribution of the mobile H_2O -accessible regions in the wood polymers. While this agrees with previous work showing that the H_2O - accessible regions of cellulose and lignin have increased mobility at higher hydration level, the elastic response here has contributions from all H-atoms. Hence, simulations could help understand how much each type of motion contributes to the average MSD.

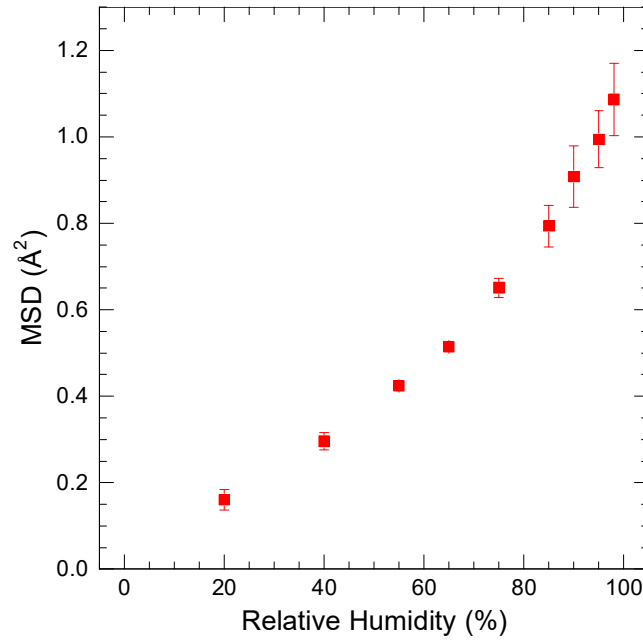


Figure 5.8 Average mean square displacement (MSD) of mobile hydrogens in the latewood sample as a function of humidity.

5.2.2 Adsorbed water dynamics and QENS broadening

Generally, the QENS signal is fitted with a model to quantify the quasielastic broadening. The most common model usually consists of the sum of a Lorentzian and a background, both convoluted with the resolution function as shown below.

$$S(q, \omega) = \{A(q)\delta(\omega) + B(q)\frac{1}{\pi}\frac{\Gamma^2(q)}{\Gamma^2(q) + \omega^2} + C(q, \omega)\} \otimes R(q, \omega) \quad \text{eq. 5.3}$$

Here $A(q)$ and $\delta(\omega)$ are the prefactor and the delta function describing the elastic intensity. The quasielastic intensity is described by the Lorentzian function with broadening Γ and prefactor B . The background term is $C(q, \omega)$ and the resolution function is $R(q, \omega)$.

However, it was found that the single Lorentzian fitted the data poorly throughout the humidity range studied, Figure 5.9 (a), and it was necessary to evaluate other models, including a stretched exponential and a two-Lorentzian model. By analyzing the residuals of these models as well as

the obtained parameters, it was determined that the two-Lorentzian model was the best fit for the data throughout all the humidity values measured as indicated by the randomness in the residuals. A comparison between the standardized residuals from the single Lorentzian and the two-Lorentzian model is provided on Figure 5.9.

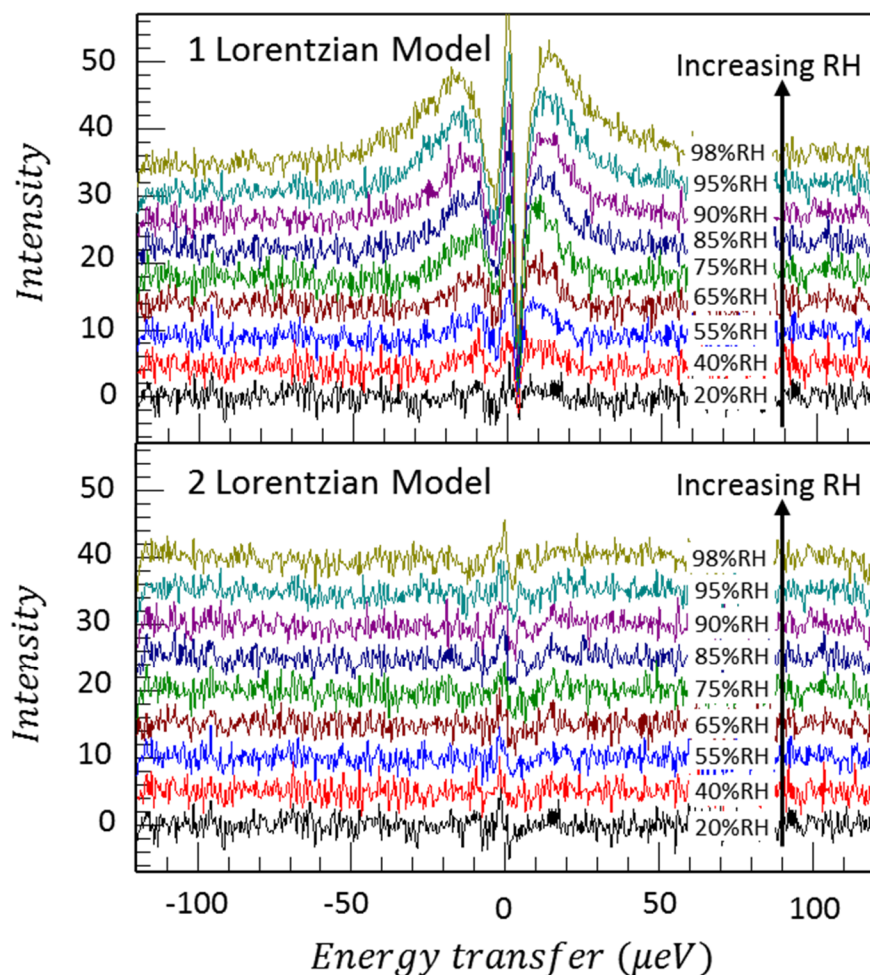


Figure 5.9 Residuals obtained from a) model with one Lorentzian and a linear background, and b) model with two Lorentzian and a linear background. For clarity purposes, each residual is offset by 5 in the y-axis.

Using this model also allowed the broadening to be quantified with only one assumption: that there were two distinct dynamic processes giving rise to the QENS signal. Furthermore, a two Lorentzian model is favored over the traditional stretched exponential models whenever there is

more than one dynamic process because it can capture the parameters describing each process rather than providing an average.¹⁹ Thus, the data at each q and RH was fitted using the following two-Lorentzian model:

$$S(q, \omega) = \{A_D(q)\delta(\omega) + L_1(q)\frac{1}{\pi}\frac{\Gamma_1(q)}{\Gamma_1^2(q) + \omega^2} + L_2(q)\frac{1}{\pi}\frac{\Gamma_2(q)}{\Gamma_2^2(q) + \omega^2} + C(q, \omega)\} \otimes R(q, \omega) \quad \text{eq. 5.4}$$

Here, the elastic intensity is described by a delta function $\delta(\omega)$ centered at approximately zero energy with area $A_D(q)$; and the quasielastic intensity is described by two Lorentzian terms, each one with a half-width half maximum $\Gamma_n(q)$ and an area $L_n(q)$. The background was fitted to a linear term $C(q, \omega)$ and $R(q, \omega)$ is the resolution function. The slower dynamics are described by the narrow Lorentzian, with area $L_1(q)$ and half-width half maxima Γ_1 , whereas faster dynamics are described by the broad Lorentzian, with area $L_2(q)$ and half-width half maxima Γ_2 . Figure 5.10 shows an example of a dataset fitted with the model.

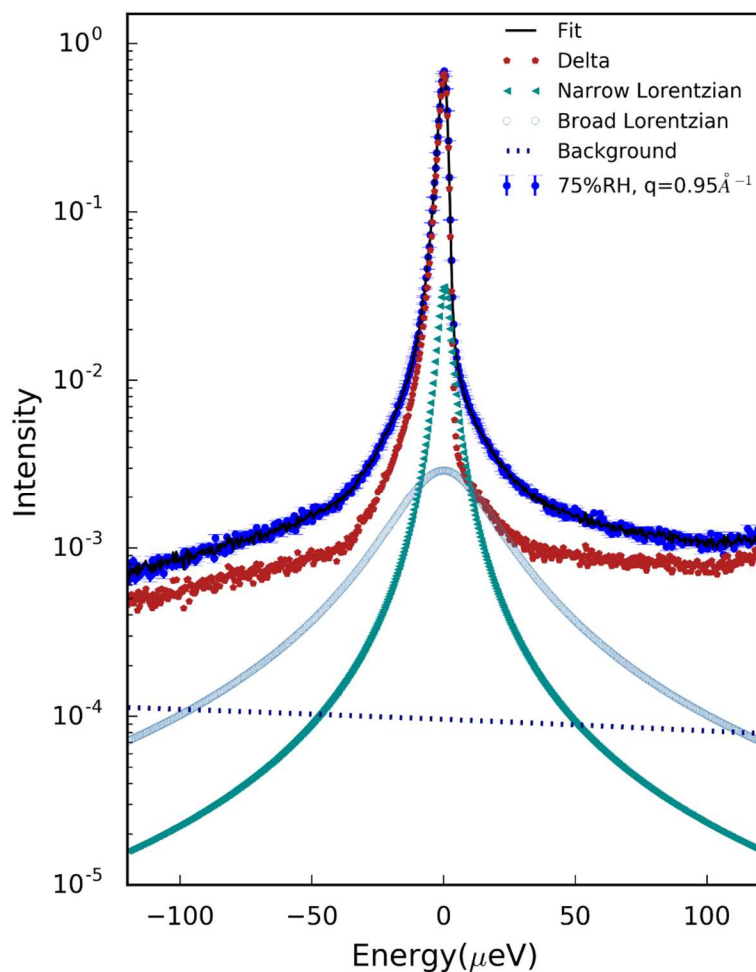


Figure 5.10 QENS spectra collected at 75% RH. Data corresponds to $q=0.95\text{\AA}^{-1}$. Fit is overlaid using a black line and individual terms of the model are also included in the plot, such as the narrow and the broad Lorentzian, and the linear background.

Within the q and ω range investigated, both components become resolution limited below 20% RH. Furthermore, the narrow component is only reliably determined above 55% RH. This suggests that below 55% RH the dynamics responsible for the narrow component are either not present or they are much slower than the dynamic range available at BASIS.

Since the moisture-induced mobility of the hydroxyl side groups lie between 1 to 100 ns,^{1,6,18} these should not contribute to the quasielastic broadening measured at BASIS. Hence, only the dynamics of the water molecules entering the cell wall are responsible for the quasielastic broadening observed in this study. Figure 5.11 shows the half-width half maxima (HWHM) of the two Lorentzians as a function of the wave vector transfer squared. The linear increase ($q < 0.5\text{\AA}^{-1}$) is characteristic of translational diffusion, whereas the flattening of the curve at higher q corresponds to spatially bound motion.

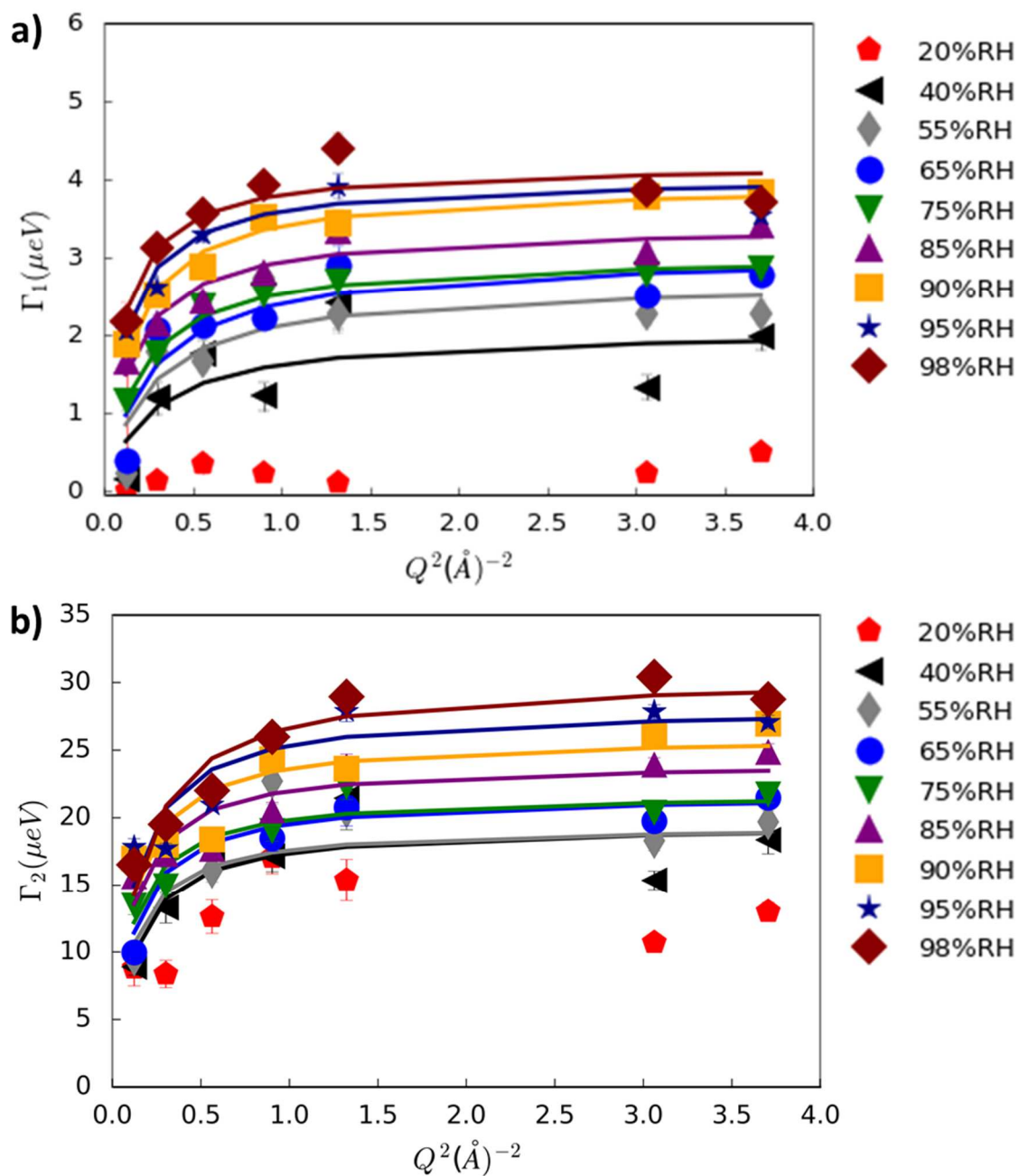


Figure 5.11 Half-width half-maxima q dependence (a) Γ_1 (slow component) and (b) Γ_2 (fast component). Filled circles correspond to the data points and overlaid lines are the fits to the jump diffusion model.

The features observed in the q dependence of the half width half maxima, shown in Figure 5.11, are characteristic of jump diffusion and can be described by the following model^{20,21}:

$$\Gamma_i(Q) = \frac{D_i q^2}{1 + D_i \tau_i q^2} \quad \text{eq. 5.5}$$

By fitting both $\Gamma_1(q)$ and $\Gamma_2(q)$ to this model, we obtained the self-diffusion coefficients (D_i) and the residence times (τ_i) of the slow and fast water as a function of RH, Figure 5.12. Each residence time is the average time between two consecutive jumps of average length l , and it is related to the translational self-diffusion coefficient according to the Einstein relation shown below.

$$D_i = \frac{\langle l^2 \rangle}{6\tau_i} \quad \text{eq. 5.6}$$

While both components are well described by the spatially bound motion characteristic of the jump diffusion model, the fast water (Γ_2) has a self-diffusion coefficient whose value is 10 times larger than the slow water (Γ_1), indicating that the fast water motion is less restricted. Highly diffusive motions, like the fast water motion, generally have short residence times, which was also observed in this study as shown in Figure 5.12. As expected from the trends in the diffusion coefficients, the residence time of the slow water is also much higher than the fast water.

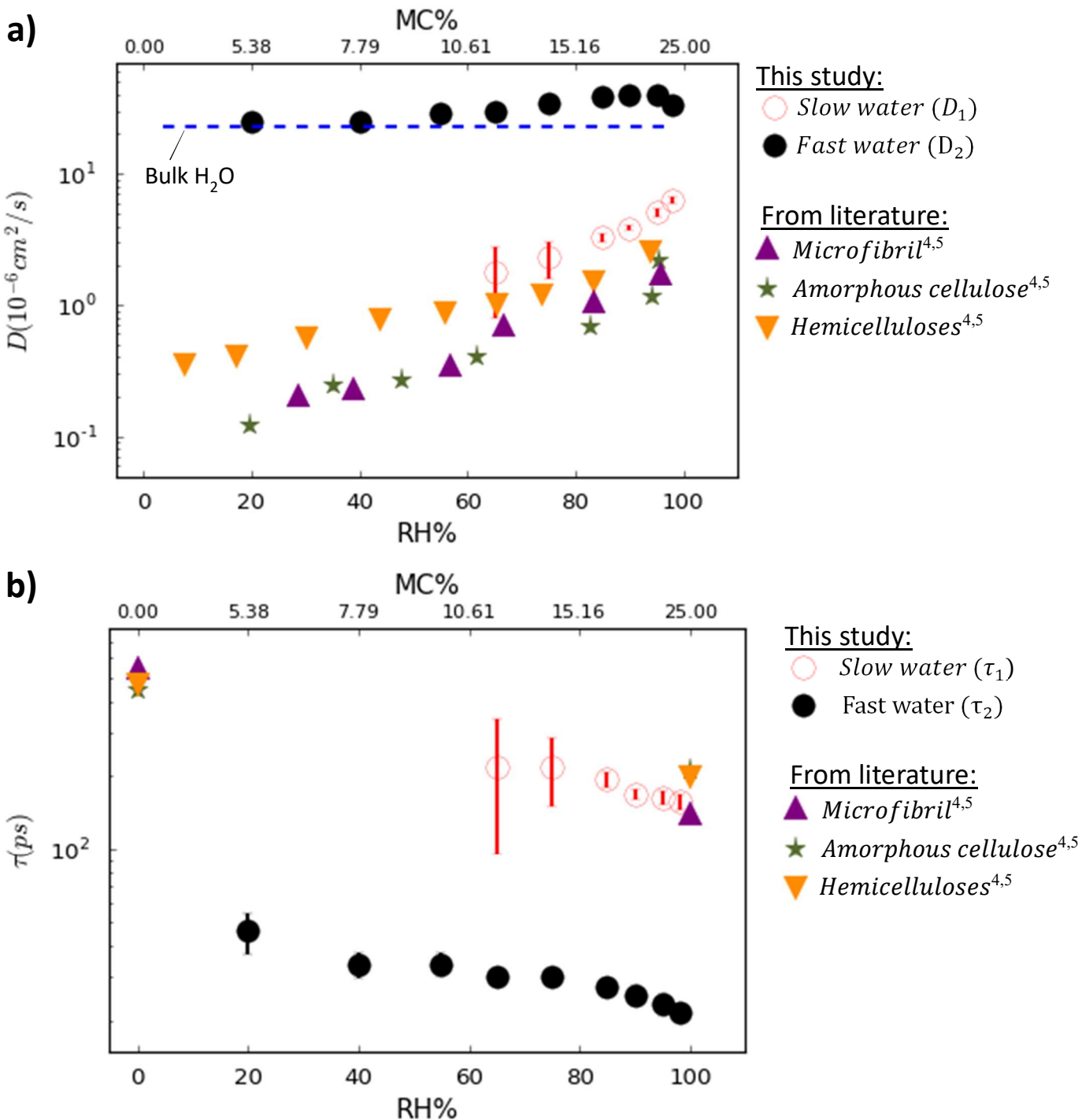


Figure 5.12 a) Diffusion coefficients and b) residence times for water absorbed in the cell wall obtained from the model. In the plot are also included values from MD simulations reported by Kulasinski et al.^{4,5} that correspond to water absorbed in amorphous cellulose, hemicelluloses and a “microfibril”, which consisted a crystalline core representative of an elementary fibril surrounded by hemicelluloses.

The humidity dependence of the fast water is most noticeable in the residence time, which tends to decrease with humidity (from 46 to 21 ps). This decrease in residence time is accompanied by a slight increase in the diffusion coefficient from 25 to $30 \times 10^{-6} \text{ cm}^2/\text{s}$. Likewise, the diffusion coefficient and residence time of the slow water also show humidity dependence above 60% RH. Below 60% RH the parameters for the slow water could not be reliably determined. Using equation 5.6, the average jump length of the slow water was found to increase from 4.7 to 7.7 Å in the range between 20% and 98% RH, whereas the fast water jump length varied between 8.4 and 6.6 Å in the same RH range.

Confined water is known to have lower self-diffusion coefficients as well as higher residence times than bulk water.^{21,22} This is consistent with the diffusion and residence times obtained for the slow water, which we attribute to water that is tightly bound to the cell wall polymers. Moreover, the magnitude of the values obtained for the slow water is comparable to the magnitude of the values reported for water absorbed by the wood cell wall polymers in simulation studies.⁵

Conversely, the diffusion coefficient of the fast water resembles that of bulk water but its residence time is much higher (~150 vs. 1 ps). This behavior has been observed in water that is trapped inside nanopores of Vycor²² or silica¹⁰ indicating that the dynamics of the fast water correspond to water that is very loosely bound.

5.2.3 Elastic Incoherent Structure Factor Analysis

The elastic incoherent structure factor (EISF), which is defined as the ratio of the elastic intensity to the total intensity, is related to the volume where the molecular diffusion occurs. At low humidity, when the elastic contribution is the highest, the EISF tends to one as expected. Figure 5.13 shows how the EISF decreases with increasing humidity, which is expected because of the increased mobility of hydrogens in the sample, particularly in the added moisture.

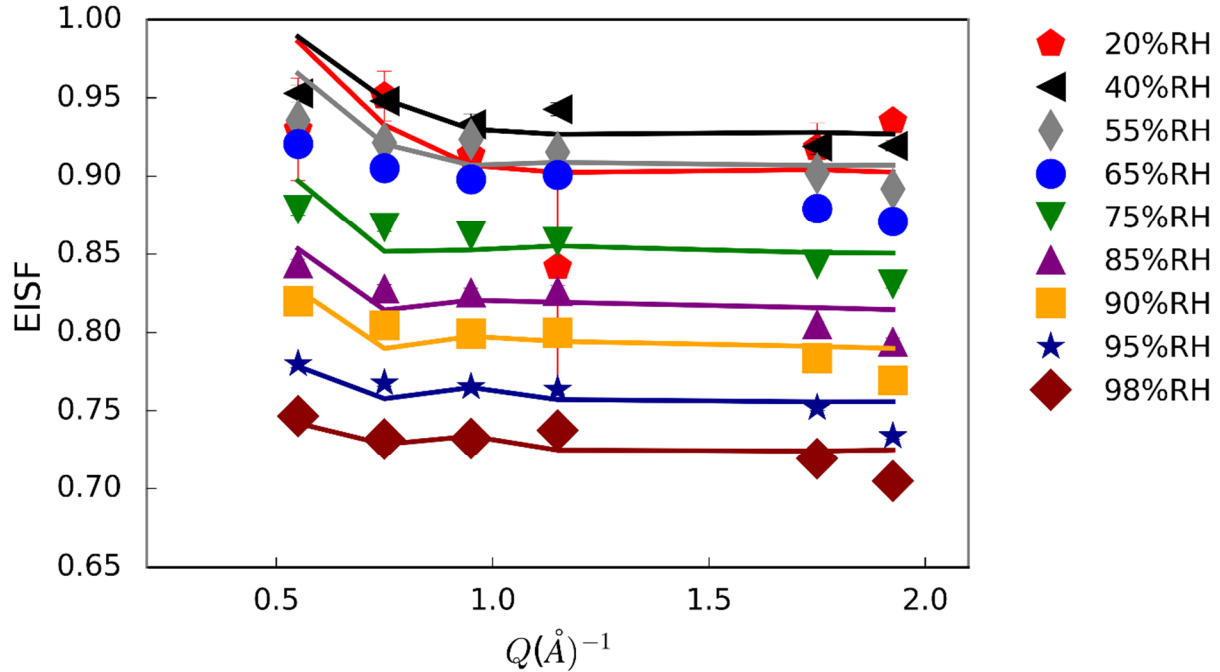


Figure 5.13 Elastic incoherent structure factor of wood as a function of humidity. Filled symbols correspond to measured data and overlaid lines correspond to the fits.

To estimate the size of the volume available for water molecules to diffuse, we assumed that diffusion occurs inside a sphere and fitted the EISF to the following model:

$$\text{EISF}(q) = c_1 + (1 - c_1) \left(\frac{3j_1(qa)}{qa} \right)^2 \quad \text{eq. 5.7}$$

where a is the confining radius, which is the radius of the sphere where water molecules can diffuse, and c_1 is the fraction of hydrogen atoms that appear immobile within the instrument resolution. In Figure 5.14, the RH dependence of the immobile fraction and the confining radius is shown. As expected the immobile fraction decreases with humidity, but this decrease becomes noticeable above 60% RH, which corresponds to the onset of the softening of the wood polymers. The radius of confinement also increases about 70% with humidity from 3.5 Å to 6 Å and like the immobile fraction most of the increase occurs above 60% RH. By comparing the obtained confining radius to the length of a OH bond (~ 0.96 Å) in a water molecule, the number of water

molecules in confinement can be estimated. Based on this approach, the estimated number of confined water molecules increases from 3 to 6 water molecules. If the estimate is based instead on the hydrodynamic radius of a water molecule, then the number of molecules would increase from 2 to 4 from 20% to 98% RH.

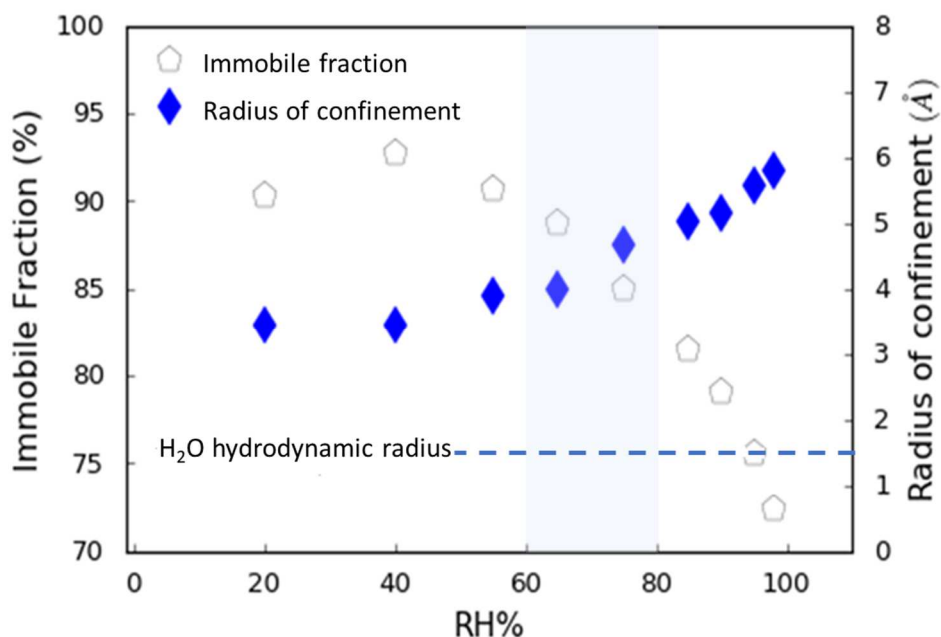


Figure 5.14 Immobile fraction of water molecules and confining radius as a function of a RH. The hydrodynamic radius of a water molecule is also shown for comparison purposes.

5.2.4 Quasielastic Incoherent Structure Factor Analysis

Similar to the EISF, we can define a quantity called the quasielastic incoherent structure factor (QISF) that can be used to determine relative contribution of each water type to the total broadening. The QISF is defined as the ratio of the area of the quasielastic intensity to the total intensity. Hence, it accounts for the intensity not accounted for by the EISF as shown below^{23–25}

$$\text{QISF} = 1 - \text{EISF} \quad \text{eq. 5.8}$$

Because in this study there are two Lorentzian terms describing the broadening of the QENS signal, there are two associated QISFs, one for each Lorentzian term. Each QISF is defined as the ratio of the area of the Lorentzian over the total area under the spectrum.

The QISF is generally fitted using form factors similar to the EISF fitting routines. This analysis allows the investigation of the size and/or shape giving rise to the broadening, but is usually used whenever the EISF analysis is inconclusive. Here, it was not possible to fit the QISF to any model. However, it can be observed that the overall intensity of both slow and fast water (QISF₁ and QISF₂) increases with humidity. Also, the peak from slow water shifts to lower q as humidity increases, Figure 5.15. This indicates that as humidity increases the size of the slow water domain also increases. A similar trend has been observed in the QISF from water trapped in silica pores,²⁴ where the shift towards lower q was attributed to an increase in size of the characteristic radius of the diffusive motion.

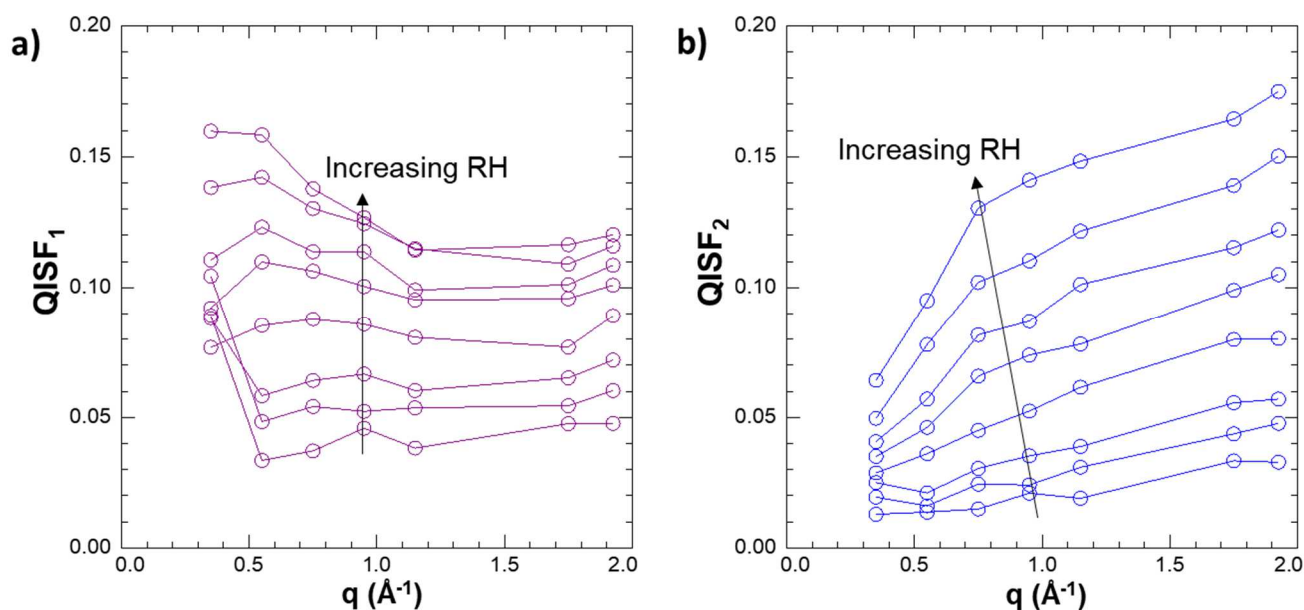


Figure 5.15 QISF q dependence for (a) slow water and (b) fast water.

To further quantify the contribution of each water type as a function of humidity, a fractional QISF (FQISF) was defined as the ratio of the QISF of each quasielastic term to the total QISF as follows.

$$\text{FQISF}_1 = \frac{\text{QISF}_1}{\text{QISF}} \quad \text{eq. 5.9}$$

$$\text{FQISF}_2 = \frac{\text{QISF}_2}{\text{QISF}} \quad \text{eq. 5.10}$$

The humidity dependence of the fractional QISF of each water type is shown in Figure 5.16. At low humidity levels, slow water accounts for most of the QISF. The fraction of slow water decreases with humidity as the fast water fraction increases. As humidity increases the fraction of fast water increases until the relative amounts of slow and fast water are the same.

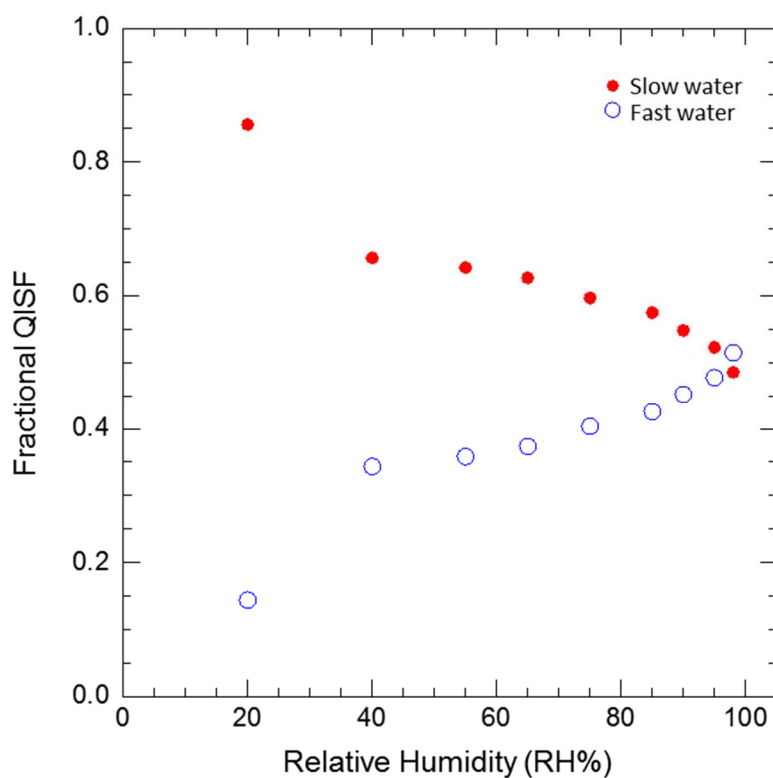


Figure 5.16 Fractional QISF of the slow and fast water as a function of humidity.

5.3 Discussion

A proposed model of moisture uptake in the wood nanostructure based on the QENS results is shown in Figure 5.17. When water enters the cell wall it can be found in two different local environments: surrounded by wood polymers or surrounded by other water molecules. At low RH, most of the water in the cell wall is slow water, meaning that it interacts strongly with the surface of the hydrophilic wood polymers. It should be noted that even though most of the water in the cell wall is slow water at low RH, the broadening caused by the slow water becomes resolution limited below 55% RH, which is why D and τ were not reported at low RH. While the magnitude of D and τ agree with water bound to the hydroxyl groups in hemicelluloses or amorphous cellulose,^{4,5} it is important to note that our results provide an average of the properties of all slow water in the cell wall. Thus, there is no distinction in the QENS signal between slow water between elementary fibrils, outside the microfibrils, or in the middle-lamella matrix. Therefore, QENS alone cannot be used to determine the distribution of slow water in the cell wall.

As RH increases and the total amount of water in the sample increases, the size of the swollen regions increases as well. The relative amount of fast water, which is water that is surrounded by other water molecules like in a water cluster, increases with increasing humidity. Although, MD simulation studies mentioned the formation of water clusters as humidity increases, neither a self-diffusion coefficient nor a residence time was reported previously for these water clusters.¹⁹ More recent MD simulation on the moisture-induced softening of the wood polymer have also provided evidence of the formation of water clusters or nanodroplets inside the cell wall above 10% MC.²⁶ The magnitude of D is similar to the values reported for bulk water, whereas the residence time is much larger than the bulk water value, which is consistent with water under confinement. Simulation studies on mobility of water in porous glasses have also reported that water near the pore surfaces has much more restricted mobility than water that is inside the pores (beyond the third layer of water molecules next to the surface).²⁷ The properties of the water inside the pores

then resemble bulk water because their mobility is no longer hindered by their interaction with the pore surfaces.²⁸ Experiments and simulations have both shown that the level of restriction on the dynamics is dependent on the pore size.²⁹

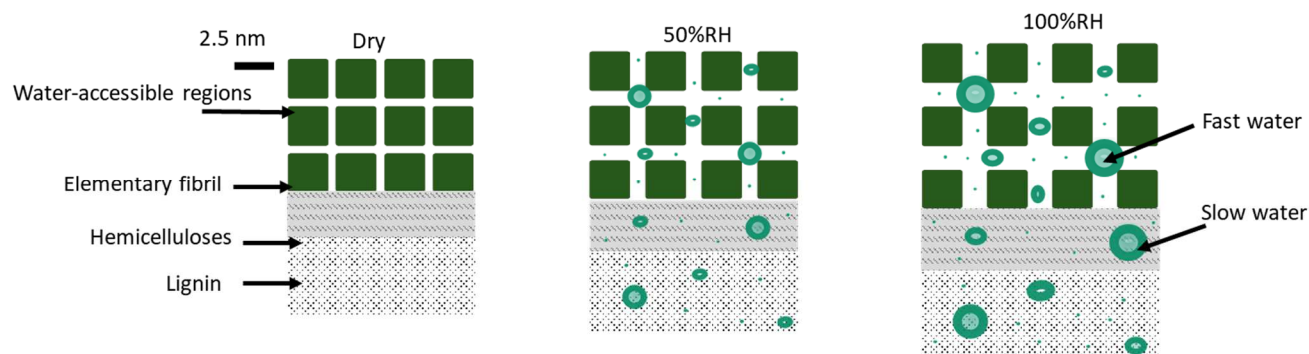


Figure 5.17 Proposed effect of moisture uptake on the S2 nanostructure. Here, the microfibril is proposed to have a rectangular cross-section of 3 by 4 elementary fibrils but only a longitudinal view of the microfibril is shown.

The idea that water within the cell wall may be found in two different local environments has been previously proposed based on NMR 2D measurements that showed that bound water had two different spin relaxation times (T_1).³⁰ While these results were attributed to water being found in two local environments, namely, less mobile water responsible for the swelling of the wood polymers and mobile water possibly found in small voids within the cell wall, no properties describing the dynamic state of each water type were reported.³⁰ In contrast, our results not only provide evidence of the existence of two water types based on the broadening of the QENS signal but also provide insight on their dynamic state.

The confining radius found from the EISF agrees the micropores whose diameter is below 1.59 nm, which according to previous NMR cryoporometry results accounts for over 70% of the total porosity in wood cell walls.³¹ The size found not only falls within the size distribution reported by NMR, but also agrees with the smaller micropores obtained via solute exclusion with the total

organic carbon technique, which showed that the mean pore size in earlywood is equivalent to a molecule with a radius of gyration of 3.77Å.³²

Even though QENS cannot provide further insight on the distribution of each water type within the cell wall, it can be combined with MD simulations to further elucidate what is the role of each wood polymer in the moisture-induced dynamics of the absorbed water. For instance, recent simulation studies find that when water molecules first enter the cell wall they occupy the empty spaces available. Then above 10% MC as the MC of the sample increases the fractional free volume of both lignin and hemicelluloses increases.²⁶ Below 10% MC, the free volume in lignin decreases. This decrease suggests that most water found in lignin at low MC will be tightly bound by the polymers and thus, contribute significantly to the total amount of slow water. Whereas, at higher MC (above 10%MC) the increase of free volume in both polymers suggests that both slow and fast water will be found in these polymers. Moreover, the relative amount of fast water in the hemicelluloses should also increase as the free volume increases. Future MD studies to simulate the QENS spectra could help determine the contribution of each water type found in each wood polymer.

5.4 Conclusions

QENS can be used to study the diffusive motions of bound water inside the cell wall. In this study, it was found that bound water exists in two dynamic states, namely, slow and fast water. Slow water corresponds to water that is near the wood polymers, whereas, fast water corresponds to confined water that is mainly interacting with other water molecules. Both types of water motion can be described by a jump diffusion model, meaning that water molecules can diffuse freely inside a free volume till they become momentarily trapped, and after some time they can diffuse freely again. By analyzing the EISF, it was found that the overall confining radius where water molecules can diffuse increases about 70% as humidity increases from 20 to 98% RH.

5.5 References

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Chapter 6. Conclusions and outlook

The first half of this chapter consists of a summary of the work presented including methods and main findings. The second half comprises an outline of the future work and implications of the results presented in this dissertation.

6.1 Synthesis

The first part of this dissertation focused on the use of SANS to study the moisture-induced swelling of nanostructures in wood cell walls as a function of humidity. The effects of sample preparation and wood source were explored. It was shown that sample preparation and selection are important and can affect the intensity and anisotropy of the scattering features observed. SANS studies on latewood unmodified specimens cut from each primary orientation allowed us to study the primary plane dependence on the SANS data, as well as the moisture-induced swelling at the elementary fibril level. Based on these studies it was found that while the anisotropic features observed at larger length scales are primary orientation dependent, the measurement of the elementary fibril swelling is not. Likewise, SANS measurements were carried on samples of two different thicknesses, namely, 500 μm and 25 μm thick. In the thinner samples, the wood cell walls were partially cut, which only resulted in higher elementary fibril spacings only at the high humidity. Finally, a comparison was made between the swelling at the S2 and elementary fibril and it was found that the majority of the swelling originates at the elementary fibril. Hence, this swelling must be addressed by wood protection treatments meant to minimize moisture-induced swelling and impart moisture-durability.

Studies on chemically modified cell walls provided further insight on how commercially relevant chemical modifications, namely PF and pMDI adhesive infiltration and acetylation, alter the cell wall nanostructure and consequently, the moisture-induced swelling at the elementary fibril level. Based on the SANS measurements obtained from samples modified with deuterium labeled

chemicals, it was found that the adhesives (PF and pMDI) infiltrated the water-accessible regions between the elementary fibrils. Acetylation, on the other hand, does not infiltrate these regions but rather modifies the surroundings of the elementary fibrils. While deuterium labeled acetylation did not result in a diffraction peak, the highly aligned scattering observed in the acetylated samples suggests that acetylation occurs preferentially in the surfaces of the lamellae formed by the microfibrils. Samples modified with hydrogenated chemicals were measured using SANS to quantify the effect of the moisture-induced swelling at the elementary fibril and it was found that only PF infiltration reduced the swelling significantly at both the elementary fibril level and the bulk level. Conversely, acetylation reduced the amount of water entering the cell wall as evidenced by the moisture-content measurements. This reduction in the moisture content in the sample led to a decrease in the humidity range where the elementary fibril peak was measurable.

QENS studies on unmodified cell walls allowed us to measure for the first time the state of the bound water in the cell wall, and revealed that water in the cell wall can exist in two dynamical states: slow and fast water. Slow water corresponds to water that is highly associated to the wood polymers, and hence, its dynamics are greatly hindered by its interaction with the wood polymers. Fast water corresponds to water that is more likely to interact with other water molecules, such as water confined inside nanopores within the cell wall. Both types of water were well described using a jump-diffusion model, which agreed with the model of water molecules whose motion can be described as a stop and go process as observed in MD simulations reported by Kulasinski et. al.¹ Moreover, analysis of the elastic intensity revealed that the confining radius of the water molecules, lies within the smallest pore size distribution reported in the literature.^{2,3}

6.2 Future work

SANS could provide valuable information on how other chemical modifications not discussed in this research, including heat treatments and the addition of preservatives, might alter the cell wall nanostructure and its moisture-induced swelling. In addition, future studies on adhesive infiltrated

cell walls could address how the amount of adhesive or its molecular weight affect the extent of adhesive infiltration into the cell wall.

With regards to analysis, an extended analysis using Fourier-transform methods to model the scattering data could aid in deconvoluting the SANS data presented in this dissertation, particularly at the low q . While 2D SLD distribution maps might not provide the level of complexity needed to replicate the anisotropic scattering patterns obtained, 3D SLD maps could. This analysis would be a valuable addition, especially to further our understanding of how chemical modifications alter the cell wall. I intend to pursue it in the future.

Future QENS studies on chemically modified cell walls could provide insight on whether wood protection treatments that impart moisture-durability have any effect on the types of bound water found in the cell wall. These future studies could provide further insight on how acetylation prevents decay. Questions like whether or not treatments are capable of preventing one of these water types from forming could help us further elucidate the nanoscale mechanisms behind moisture-durability.

6.3 References

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Appendix A: Neutron Scattering Sample Environments

The humidity chambers were developed and built in collaboration with the Forest Products Laboratory and the Oak Ridge National Laboratory instrument scientists and staff, to enable in situ humidity controlled neutron scattering experiments.

A-1. SANS Humidity Chamber

The chamber has an aluminum body and 10 quartz windows, where samples can be placed. Samples can be held in place using tape or another adhesive. Each window is 19 mm in diameter, allowing for larger beams (up to a sample aperture of 14 mm). The chamber sits on the translation stage, which allows automated data collection on all samples inside the chamber. The neutron path length inside the chamber is ~6 mm and height inside the chamber is ~22 mm. The humidity inside the chamber is controlled with a RH generator and a feedback loop control that monitors the humidity using a probe that sits in the chamber. The humidity range accessible at room temperature is between 3% and 98% RH. Above 95% RH condensation can occur in the windows at low room temperatures. While the reservoir connected to the RH generator can be filled with either H₂O or D₂O, the use of pure D₂O is recommended because even small amounts of H₂O in the internal reservoir can contribute to large amounts of incoherent scattering. Figure 3 shows an example of how the coherent scattering signal can be compromised by having H₂O only in the internal reservoir.

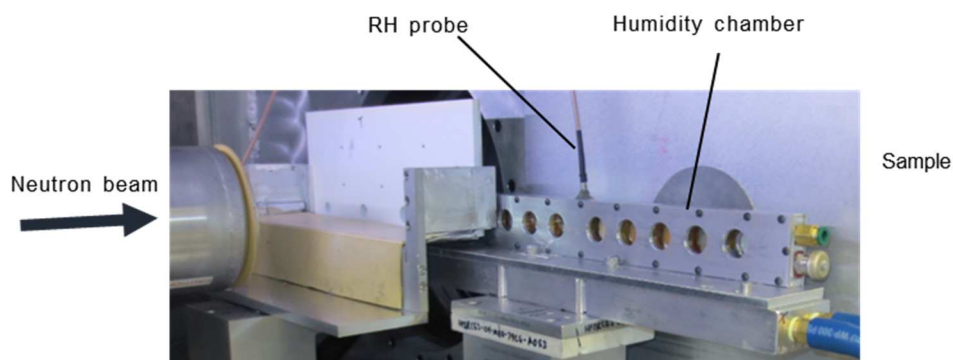


Figure A- 1 Humidity chamber already assembled and fixed onto the translation stage at Bio-SANS.



Figure A- 2 Humidity chamber showing samples held in place using polyamide tape.

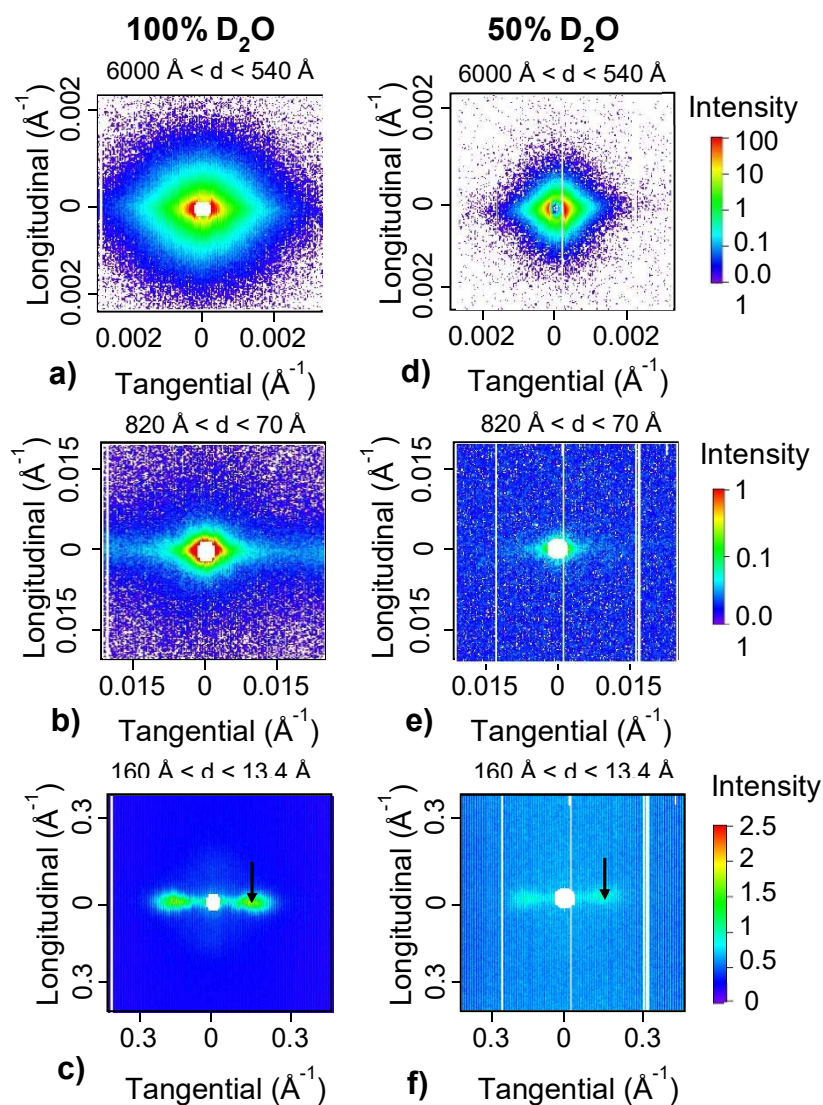


Figure A- 3 Two-dimensional anisotropic scattering patterns of 0.5-mm-thick wood sections over a broad q range from the tangential-longitudinal orientation. Samples were conditioned at 89% RH using the RH generator connected to (a-c) an external reservoir filled with pure D₂O and (d-f) an external reservoir with

D₂O and an internal reservoir with 50% H₂O. The white spot in the center of detector image is the shadow of the beam stop.

A-2. QENS Humidity Chamber

The chamber is entirely made from aluminum, the thickness of the aluminum used in each window does not exceed 2 mm. Samples can be fastened in placed using the built-in aluminum plates and screws. Each square window is 100 mm across, allowing for the full size of the incoming beam (3 - 4 cm). The chamber can be attached to the stick stage, which makes this setup compatible with all other instruments that use the stick stage at SNS. The neutron path length inside the chamber is ~6 mm and height inside the chamber is ~22 mm. The humidity inside the chamber is controlled with a LS&C Sciences RH generator (FL) using the dew point temperature method. The relative humidity and temperature inside the chamber can be monitored using a Sensirion probe that sits in the chamber. The humidity range accessible at room temperature is between 3% and 98% RH.

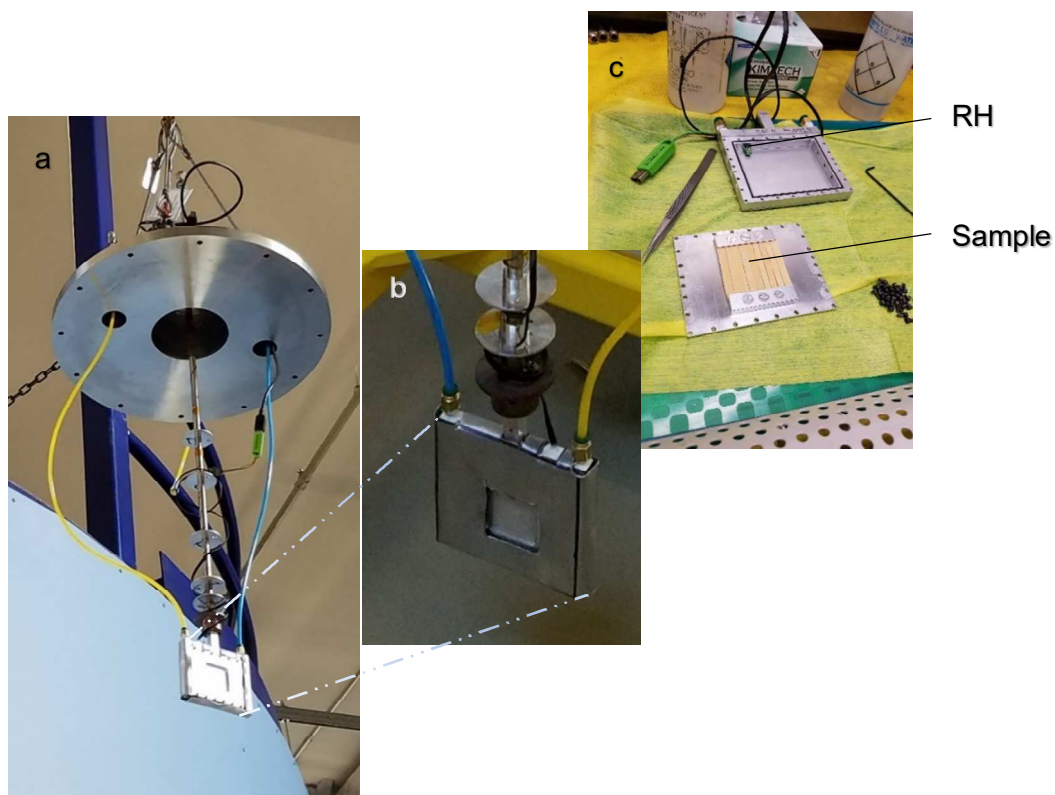


Figure A- 4 (a) Full view of the setup showing the sample chamber mounted on stick holder. (b) Close-up view of the assembled chamber with the cadmium mask used during the measurements. (c) Open chamber, where the humidity probe and the fastened sample are visible.

Appendix B: SANS Data reduction and averaging

In this appendix, all the necessary corrections that are implemented during the standard reduction of SANS data from 2D to 1D profiles will be discussed in detail. These corrections were performed using the Igor Pro Spice Procedure provided at Bio-SANS and described in detail in the following paragraphs. Note that any corrections necessary for time of flight instruments will not be discussed in this appendix.

B-1. Data corrections

An uncorrected SANS 2D detector image from a 0.5 mm-thick tangential-longitudinal latewood loblolly pine section is shown in Figure B- 1. Each pixel in the image represents a detector and the pixel value is the number of neutrons counted by the detector during the experiment. Before interpretation a series of data corrections must be applied to the images. These corrections include removing the detector dark current, normalizing to the incident beam, accounting for differences in pixel sensitivity, applying a solid angle correction, applying a transmission correction, removing background contributions, and applying absolute scaling.

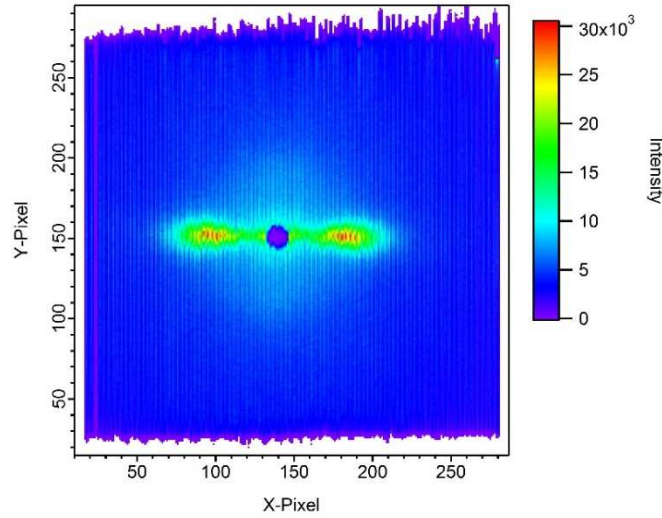


Figure B- 1 Uncorrected scattering pattern obtained at Bio-SANS from a tangential-Longitudinal sample held at 89% RH. Data was collected at a sample to detector distance of 0.3 m with a wavelength of 6Å and four guides.

Dark current primarily arises from the electronic noise in the detector, but stray neutrons that enter the detector from sources other than the neutron beam may also contribute. The dark current image shown in Figure B- 2 was obtained with the shutter closed. The sample intensity $I_s(x,y)$ is corrected for dark current using

$$I'_s(x,y) = I_s(x,y) - \frac{t_s}{t_{dc}} I_{dc}(x,y) \quad \text{eq. B.1}$$

where $I'_s(x,y)$ is the dark current corrected intensity, $I_{dc}(x,y)$ is the dark current intensity, t_s is the counting time for the sample experiment, and t_{dc} is the counting time for the dark current experiment. Dark current is assumed to be constant with time and the dark current correction is normalized using the ratio of counting times. Figure B- 3 shows the data after the dark current correction has been performed. For these measurements $I_s(x,y) \gg I_{dc}(x,y)$ and the dark current correction does not have a noticeable effect.

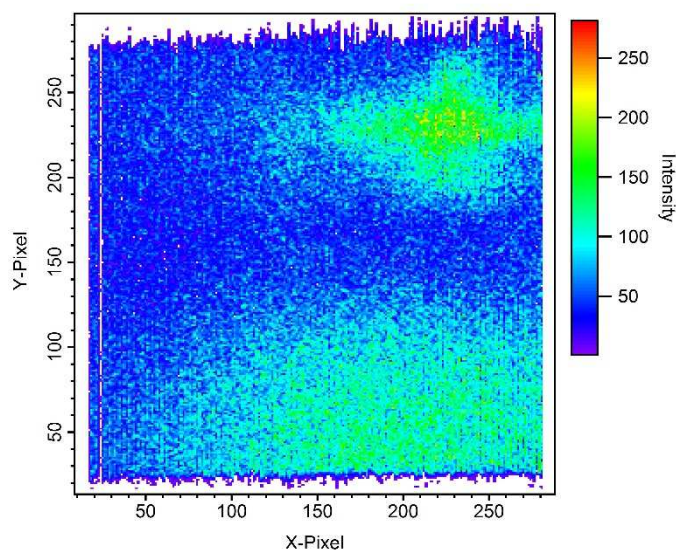


Figure B- 2 Dark current measurement obtained at a sample to detector distance of 0.3 m, using a wavelength of 6Å and four guides.

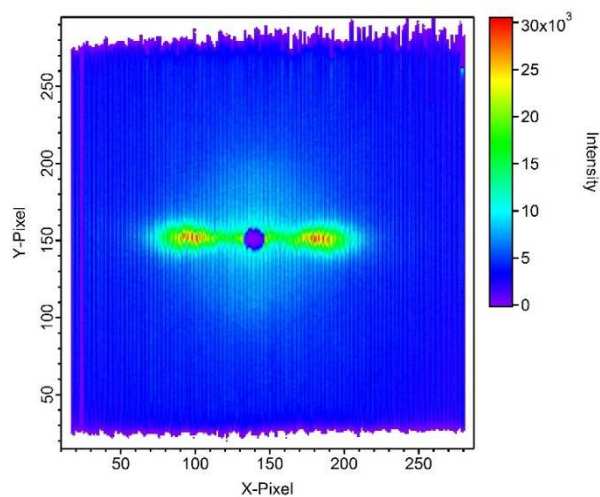


Figure B- 3 Data obtained at a sample to detector distance of 0.3 m, using a wavelength of 6Å and four guides after dark current correction.

Data normalization is performed to account for variations in neutron beam strength. The detector images are normalized by dividing the image by the number of monitor counts measured during

an experiment. The total number of monitor counts for Figure B- 3 was 7027644 and Figure B- 4 shows the normalized image.

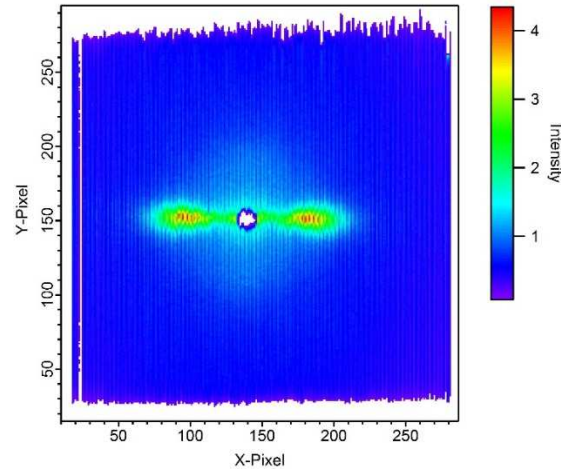


Figure B- 4 Data obtained at a sample to detector distance of 0.3 m, using a wavelength of 6Å and four guides after dark current correction, and normalization with respect to monitor counts.

A detector efficiency correction is applied to scale every data point to that of a detector of uniform efficiency. The detector efficiency is determined by measuring the scattering intensity $I_{flood}(x,y)$ of a high absorption cross-section sample whose scattering pattern is independent of the scattering angle (Figure B- 5). The dead pixels and the pixels near the edge of the detector are masked in Figure B- 6, and the detector efficiency $\varepsilon(x,y)$ is calculated using

$$\varepsilon(x,y) = \frac{n}{\sum_{x,y} I_{flood}(x,y)} I_{flood}(x,y) \quad \text{eq. B.2}$$

where n is the number of pixels. Then the detector efficiency corrected intensity is

$$I''_s(x,y) = \frac{I'_s(x,y)}{\varepsilon(x,y)} \quad \text{eq. B.3}$$

Figure B- 5 shows the normalized data corrected for pixel sensitivity and dark current.

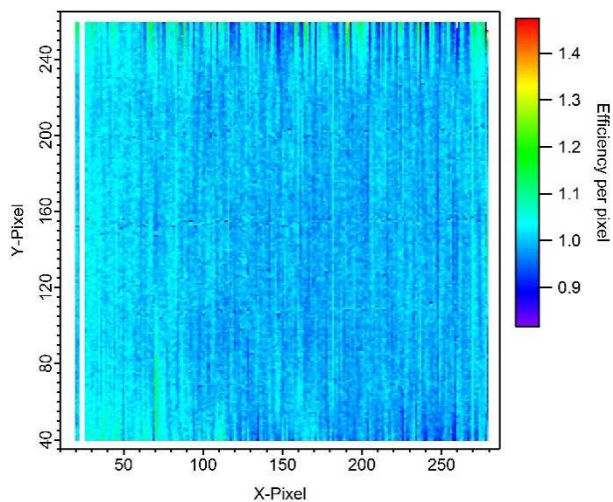


Figure B- 5 Incoherent scattering pattern from a thick plate of polymethylmethacrylate (PMMA) used to quantify pixel efficiency.

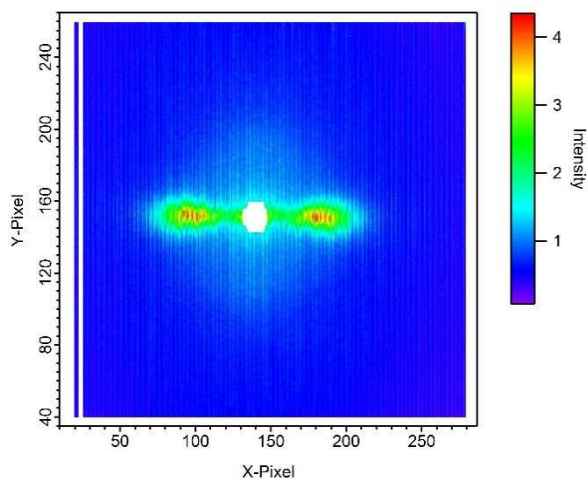


Figure B- 6 Data obtained at a sample to detector distance of 0.3 m, using a wavelength of 6Å and four guides after dark current correction and normalization with respect to monitor counts. Dead pixels, pixels near the edges and beamstop have been masked.

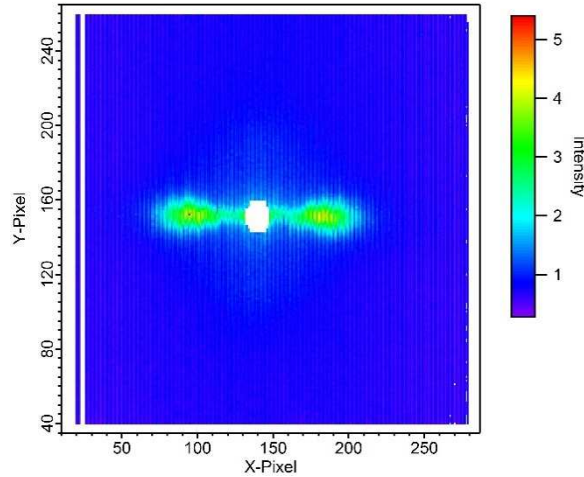


Figure B- 7 Data obtained at a sample to detector distance of 0.3 m, using a wavelength of 6Å and four guides after dark current correction and normalization with respect to monitor counts. Dead pixels, pixels near the edges and beamstop have been masked and pixel sensitivity correction has been performed.

Scattering geometry is generally represented by an Ewald sphere. However, experimentally the data is collected by a planar 2D detector (see Figure B- 8) and a solid angle correction is required.

The corrected macroscopic scattering cross section $\frac{d\Sigma}{d\Omega'}$ is calculated as

$$\left(\frac{d\Sigma}{d\Omega'}\right)_{corrected} = \left(\frac{d\Sigma}{d\Omega}\right)_{measured} \left(\frac{d\Omega}{d\Omega'}\right) \quad \text{eq. B.4}$$

where $d\Omega$ and $d\Omega'$ are the measured and corrected solid angles, respectively. A solid angle is defined as

$$\Omega' = \frac{A'}{r'^2} \quad \text{eq. B.5}$$

where A' is the surface area of the projection onto a sphere surface, and r' is radius of the sphere.

Using equation B.5 the solid angle correction factor $d\Omega/d\Omega'$ is expressed as

$$\frac{d\Omega}{d\Omega'} = \frac{A}{r^2} \frac{r'^2}{A'} \quad \text{eq. B.6}$$

where r is the vector that extends from the center of the Ewald sphere to the projected area on the detector A . Using trigonometry r and r' are related by

$$\frac{r'}{r} = \cos \theta \quad \text{eq. B.7}$$

and A and A' by

$$\frac{A}{A'} = \cos \theta \quad \text{eq. B.8}$$

Then substituting equations 7 and 8 into equation 6, $d\Omega/d\Omega'$ is expressed as

$$\frac{d\Omega}{d\Omega'} = \cos^3 \theta \quad \text{eq. B.9}$$

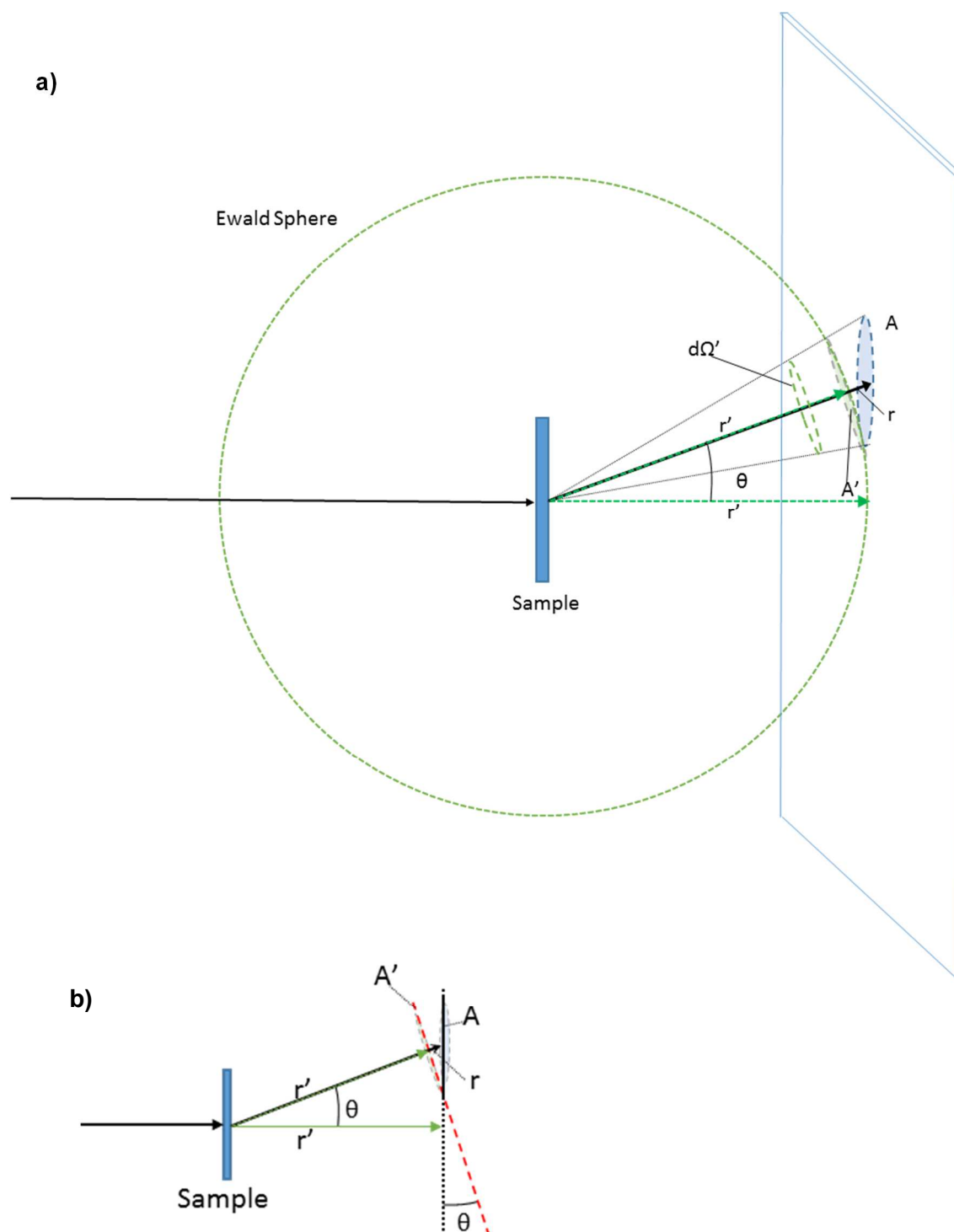


Figure B- 8 (a) Schematic of the scattering geometry showing the Ewald sphere and the solid angles. (b) Close-up on the scattering geometry showing the scattering angles and the projected areas

The fall of a neutron due to gravity after it is scattered by the sample Δy must also be taken into account using

$$\Delta y = \frac{1}{2} * g \left(\frac{m_N * SSD * \lambda}{h} \right)^2 \quad \text{eq. B.10}$$

Here g corresponds to the acceleration due to gravity, and λ is the neutron beam wavelength.

Transmission is the portion of the incident beam that passes through the sample without scattering. It is measured by taking the ratio of the beam intensity measured by the detector with and without a sample in the beam. The beam must be attenuated to prevent detector damage. For the sample to detector distance of 6m, transmission measurements were taken simultaneously with sample scattering data by using a semitransparent beamstop. An example of the experiments used for transmission are shown in Figure B- 9. The 6 m transmission measurements were used for correcting both 6 m and 0.3 m SSD's. At 14.5 m SSD transmission measurements were taken separately because there was not a semi-transparent beamstop available for this configuration and the upstream attenuator between the velocity selector and beam monitor was employed. Using the transmission measurements, the transmission correction factor is calculated as

$$T = \frac{\sum_x \sum_y \frac{I_s(x,y)}{m_s}}{\sum_x \sum_y \frac{I_{empty}(x,y)}{m_{empty}}}; \text{ where } \Delta x, \Delta y < R \quad \text{eq. B.11}$$

where m_s and m_{empty} beam are the monitor counts for the sample and empty measurements, respectively. Only the intensity of the pixels within the beam radius R from the beam center were used for calculating T .

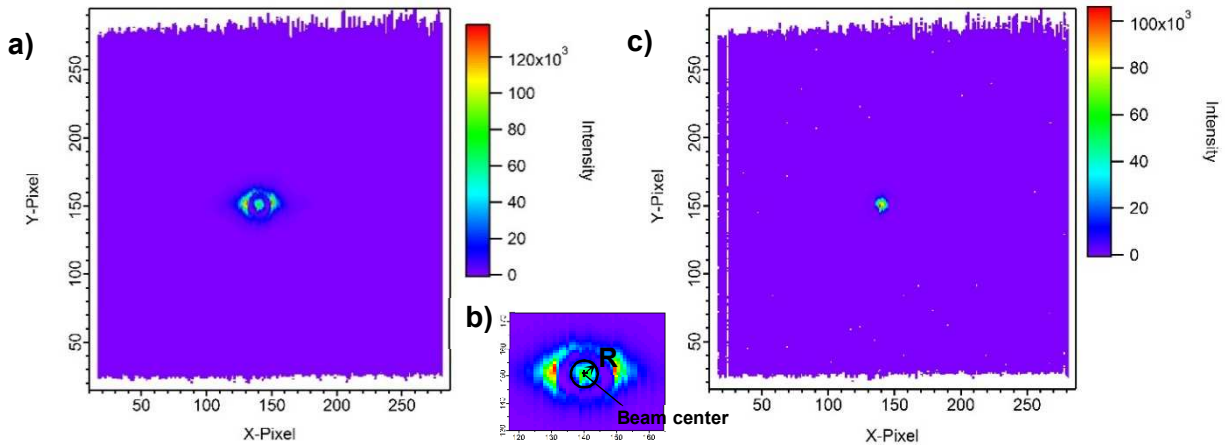


Figure B- 9 (a) Transmission measurements of tangential-longitudinal sample held at 89% RH. (b) Close-up on the transmission measurement of a tangential-longitudinal sample at 89% RH showing the beam center and a circle of radius R which contains the pixels used for calculating the transmission coefficient. (c) Transmission data of the empty beam at 89% RH (without samples or sample chamber). Both measurements were taken at a sample to detector distance of 6m, using a wavelength of 6Å and four guides.

Then, the corrected intensity I''' is calculated using T using

$$I'''(x, y) = \frac{I''(x, y)}{T[1+\sec(2\theta)]/2} \quad \text{eq. B.12}$$

Figure B- 10 shows the 2D scattering pattern corrected for transmission.

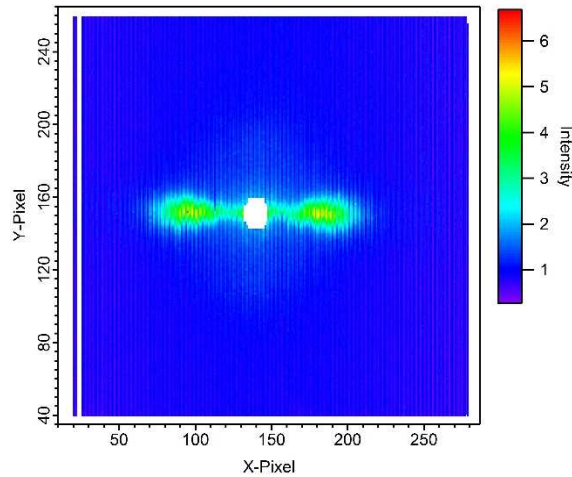


Figure B- 10 Data obtained at a sample to detector distance of 0.3 m, using a wavelength of 6Å and four guides after dark current correction and normalization with respect to monitor counts. Dead pixels, pixels

near the edges and beamstop have been masked and pixel sensitivity correction has been performed. Data has also been corrected for transmission using the empty-beam transmission method.

External background contributions such as scattering from the sample holder, as well as scattering from the environment including RH contributions and stray neutrons, were taken into account by measuring the scattering data of an empty cell, Figure B- 11, at every RH step for each instrument configuration. Background data was normalized and corrected for dark current, transmission and dead pixels and then subtracted from the uncorrected data set.

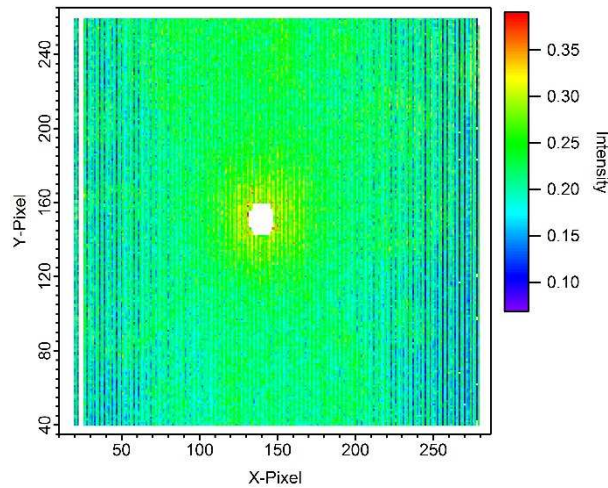


Figure B- 11 Background scattering at 89% RH, data was obtained by measuring the empty cell at a sample to detector distance of 0.3m using a wavelength of 6Å and four guides after dark current correction and normalization with respect to monitor counts. Dead pixels, pixels near the edges and beamstop have been masked and pixel sensitivity correction has been performed. Background data has also been corrected for transmission using the empty-beam transmission method.

The data was then transformed from arbitrary units to absolute to obtain the macroscopic scattering cross-section using

$$I''''(x, y) = \frac{I'''(x, y)K}{d} = \left(\frac{d\Sigma}{d\Omega} \right)_{sample} \quad \text{eq. B.13}$$

where d is sample thickness and K is the absolute scaling factor calculated using a standard sample (STD) with known scattering cross section at $q=0$. Note that K must be measured at each

instrument configuration used in the sample experiments. Here K was assessed using a porous silica oxide standard called Porasil C3 and the scattering patterns are shown in Figure B- 12. Then, the absolute scaling factor K was calculated as

$$K = \frac{d_{STD} * T_{STD+cell}}{I(Q=0)_{STD}} \left(\frac{d\Sigma}{d\Omega} \right)_{STD} \quad \text{eq. B.14}$$

Where $I(Q=0)$ corresponds to the intensity at $Q=0$, d_{STD} is the thickness of the standard, $\left(\frac{d\Sigma}{d\Omega} \right)_{STD}$ is the macroscopic scattering cross-section, and $T_{STD+cell}$ is the transmission of the standard held in the sample holder used for all the other measurements. A fully corrected data set is shown in Figure B- 13.

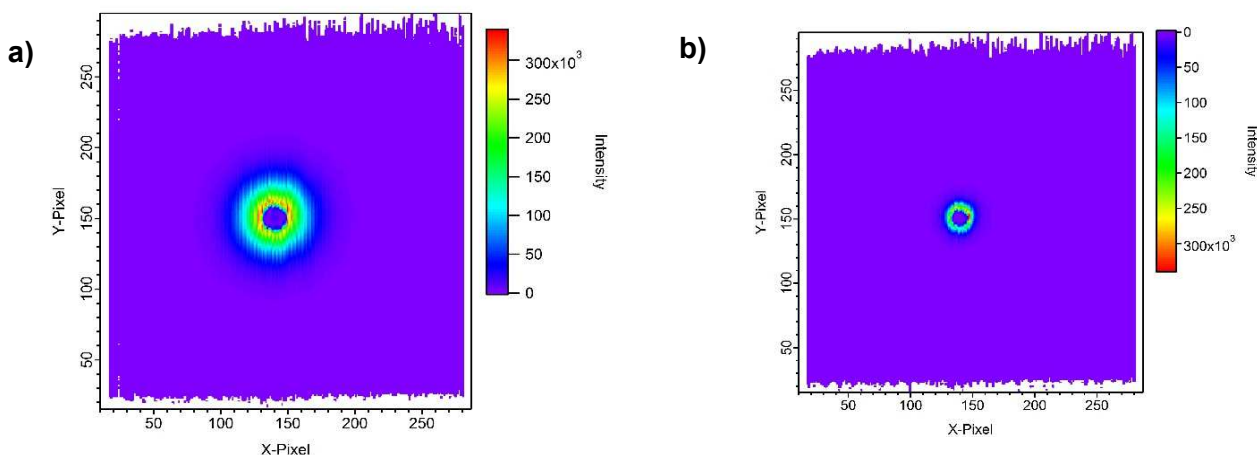


Figure B- 12 Isotropic scattering pattern of Porasil C3 obtained using a wavelength of 6Å with four guides at a sample to detector distance of (a) 6m and (b) 0.3 m.

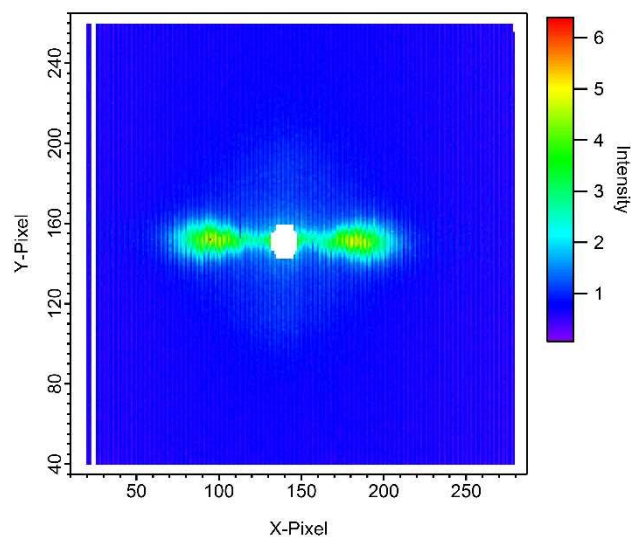


Figure B- 13 Fully corrected and scaled scattering pattern from a tangential-longitudinal 0.5mm thick section held at 89% RH. Measurement was taken at a sample to detector distance of 0.3 m using a wavelength of 6Å and four guides.

B-2. Data averaging methods: From 2D to 1D

Once the 2D data has been corrected and scaled, it is reduced to 1D. Circular binning is the standard for averaging isotropic scattering data (Figure B- 14). Whenever anisotropy is observed the circular binning is no longer appropriate and sector or rectangular averaging must be used. Generally, sector averaging is preferred over the box method because even though both enhance the anisotropic features using the latter might result in smearing the data along the length of the box by neglecting the incoherent background contribution. Hence, here we used the sector averaging method, Figure B- 15. In this method, the incoherent scattering contribution is included by using an adequate sector size, Figure B- 15(a). However, varying the sector size does not only enhance or decrease the intensity, it can also shift the Bragg peak position, Figure B- 15 (b).

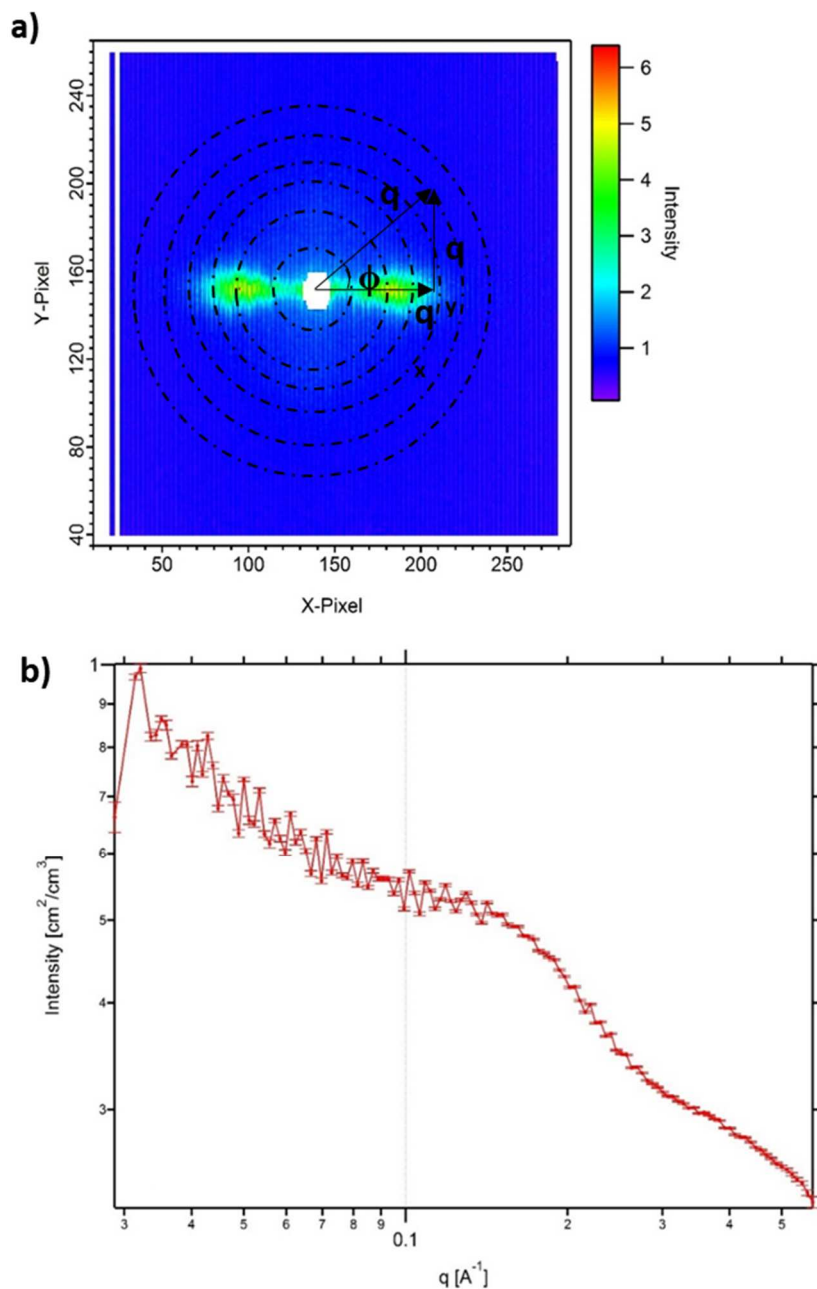


Figure B- 14 a) Circular binning performed on the scattering pattern of a tangential-longitudinal 0.5 mm thick loblolly pine latewood specimen held at 89% RH. (b) Linear profile obtained from the circular binning, note how the anisotropic features are washed out. Data acquired at Bio-SANS at a sample to detector distance of 0.3 m with a wavelength of 6Å and four guides.

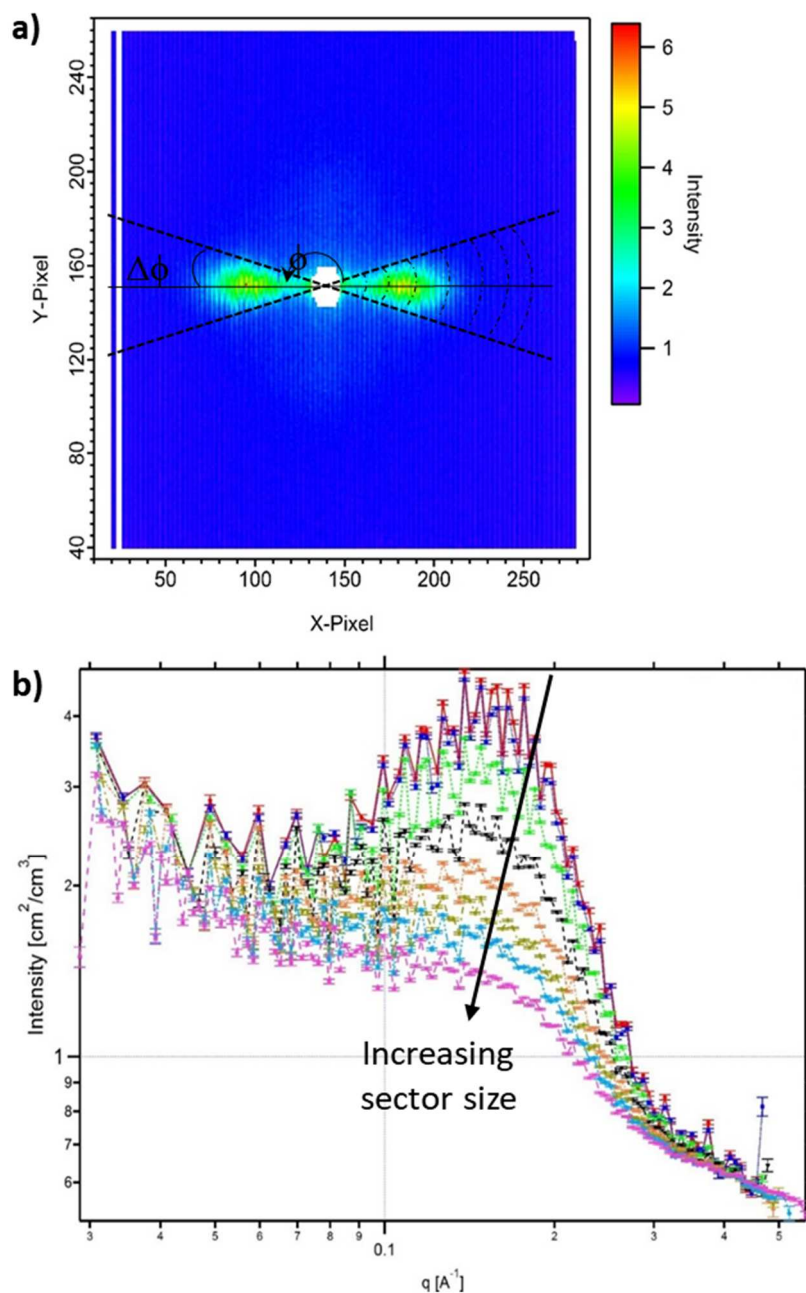


Figure B- 15 (a) Sector averaging method performed on the scattering pattern of a tangential-longitudinal sample a tangential-longitudinal 0.5 mm thick loblolly pine latewood specimen held at 89% RH. Data was acquired at Bio-SANS at a sample to detector distance of 0.3 m with a wavelength of 6Å and four guides. (b) Linear profile obtained from the sector averaging. Note how the overall intensity and the apparent position of the Bragg peak change as a function of the sector size.