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Confocal Raman Spectroscopy – Applications on Wood, Pulp, and Cellulose Fibres

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Summary: The manifold possibilities using dispersive Raman spectroscopy along the production chain from wood to cellulose fibre are presented here: The distribution of the lignin, cellulose, and added resins in the wood cell; additives inside regenerated cellulose fibres; their accessibility and reactivity. On-line measurements during the phase transition of magnesium sulphite - hexahydrate into -trihydrate are performed in a stirring vessel.

Keywords: cellulose; melamine, phase transition; Raman spectroscopy; wood

Introduction

In the last 15 years Raman spectroscopy has been used mainly in academic research. Appearance of fluorescence, which masks the Raman spectra due to it's higher intensity (up to 10^6 times) was one of the major reasons for the restricted applications. Because most fluorescent compounds have no electronic absorption bands in the near infrared, FT-Raman is preferably used with a 1064 nm light source. A laser with 785 nm, however, can combine a fluorescence prevention with a high spatial resolution of the dispersive Raman spectroscopy in comparison to the FT-Raman.[1]

Particularly the investigation of wood samples needs sufficient fluorescence suppression and prevention. Here, the Raman micro-spectroscopy is a promising technique to study lignin and cellulose simultaneously[2]. The obtained vibrational spectra of the wood samples contain contributions from all its major constituents. The carbohydrate polymers, cellulose and hemicelluloses, have similar σ-bond frames (arrangements of single bonds), and thus their spectra are expected to be similar[3]. Lignin, however, represents a heterogeneous polymer with π-contributions (aromatic structures) and produces different spectra. The vibrational information is combined with the spatial resolution by applying confocal Raman spectroscopy. In this technique[4] the excitation laser is focussed through a microscope objective on a small area (spot size 2 μm) of the sample. The detected Raman

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scattering signal is limited by a pinhole to the area surrounding the focus. The size of the pinhole determines the depth resolution, the smaller the pinhole the higher the resolution, but at the expense of the signal intensity.

Additionally, on-line, in-line, and at-line process monitoring can be accomplished by the connection of fibre optics between laser, microscope, and detector.

One example for on-line measurements is the monitoring of magnesium sulphite hexahydrate-trihydrate phase transition[5] in the SO_2 recovering cycle of the acid magnesium bisulphite pulping process. One part of the complex recovery system is an absorption unit, where the SO_2 is removed from the waste gas of the thick liquor burner by reacting with $Mg(OH)_2$ to magnesium bisulphite. The magnesium bisulphite, which is re-introduced in the pulping process as cooking liquor is formed in a two-step reaction. A major problem during the absorption recovery process is the precipitation of the solid magnesium sulphite hydrates $MgSO_3 \cdot xH_2O$. These solids cause several problems, such as mechanical abrasion, incrustation and higher loss of chemicals by the discharge of solids. Markant's[6] conclusions state that only dissolved magnesium sulphite undergoes the absorption reactions. Therefore, the kinetic of the precipitation becomes an important factor for the operation and construction of the absorption recovery system.

Two different types of magnesium sulphite hydrates are formed under process conditions, magnesium sulphite hexahydrate ($MgSO_3 \cdot 6H_2O$) and trihydrate ($MgSO_3 \cdot 3H_2O$). The two hydrate types differ in their solubility, which was first reported by Hagisawa[7]. Due to the smaller solubility of the trihydrate, process conditions should be used, which prohibit the transition to the hexahydrate. The stable solid phase is magnesium sulphite hexahydrate at temperatures below 42°C, whereas the trihydrate is the stable solid phase above 42°C. However, the metastable hexahydrate mainly crystallises at temperatures above the transition temperature of 42°C.

Experimental

Wood: Samples (measuring about 5 mm x 5 mm x 10 mm) of beech (*Fagus sylvatica*) sapwood from the same stem section were taken without dehydration and embedding, freshly cut with a sliding microtome (Reichert) into 10 µm sections, and stored in a water-glycerol mixture. The wet sections were transferred to glass microscope slides, washed with deionised water, dried in air, and fixed with an adhesive tape to avoid motion during the measurement. The Raman analysis was performed with a HoloLab Series 5000

Modular Raman Spectrometer (HL5R) from Kaiser Optical Systems Inc. (USA) equipped with f/1.8 optics, transmission grating, multichannel CCD array detector (optimised for NIR), and a 785 nm diode laser (400 mW) coupled via single mode fibre to the microscope (approximately 10 mW on the sample). A confocal fibre with a pinhole of 20 µm was used between the microscope and the detector. The lateral resolution was about 1-2 µm using the 100X objective.

Wood melamine composites: The samples (30mm x 12mm x 10mm (radial x tangential x longitudinal) were prepared from spruce (*Picea abies L. [Karst.]*). Water saturation was followed by a dehydration with methanol (4x2 hours), the resin infiltration was done simply by immersing the sample into the melamine (MER, AMI™), which was dissolved with methanol at a weight percentage of 50:50. After 3 days of immersion the samples were dried overnight at 60°C and cured at 180°C for 30min. 10µm thick transverse sections were cut from the same sample by a rotary microtome for the Raman microscopy. The sections were transferred to glass microscope slides and fixed with an adhesive tape. The measurements were done with the spectrometer described above, despite the use of a more powerful 785 nm laser (500 mW, Toptica Photonics AG, Martinsried, Germany) with approximately 80 mW on the sample.

Fibre cross sections: A fibre bundle was pulled through a 1 mm pinhole in a metallic plate of the same size as the microscope slide and cut with a razor blade.

Online measurements: The experimental setup for the online measurements of the transition kinetics from magnesium hexahydrate to trihydrate is shown in Figure 1. Into a double-jacketed, temperature-controlled vessel, 350 mL water and 40 g MgO (Fluka 63091, different kinds of MgO used in the recovery process) were agitated. The SO_2 gas was introduced into the vessel until the desired starting pH-level (between 6 and 8) was reached. The gas flow was stopped and the solution was stirred at the defined temperature (55°C, 60°C, 65°C, and 70°C) until the end of the transition. The formation of the different hydrate types was monitored online by Raman spectroscopy. A NIR immersion sampling optic (1/4") with short fixed focus on the sapphire window was connected to the holographic filtered probe head Mark II. The measurements were performed with approximately 120 mW on the sample. The pH was determined with a pH electrode linked to a Metrohm E588 pH-meter.

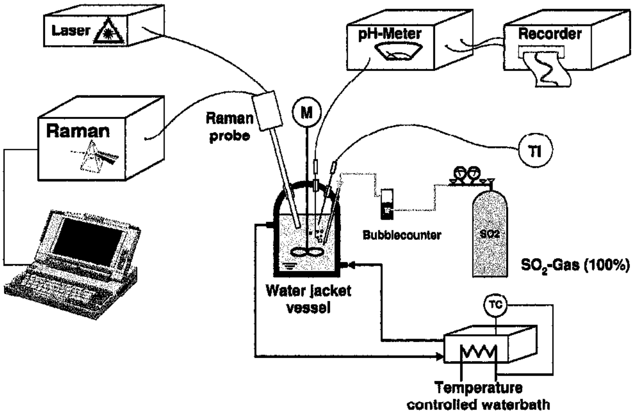

Figure 1. Experimental setup for the online measurement of the phase transition kinetics of magnesium sulphite hydrates by Raman spectroscopy.

Results and Discussion

The resolution of the confocal Raman spectroscopy is limited by the diffraction depending on the wavelengths. A 785 nm laser was used to avoid fluorescence. The distribution of the lignin and the cellulose in the individual cell wall layers of wood samples could be determined. The lignin content was identified by the signal around 1602 cm^{-1} (peak area between 1593 cm^{-1} and 1614.5 cm^{-1}) representing the stretching of the aromatic ring and the cellulose content was determined by the signals at 1092 cm^{-1} and 1123 cm^{-1} (peak area between 1074 cm^{-1} and 1134 cm^{-1}, Figure 2)[8].

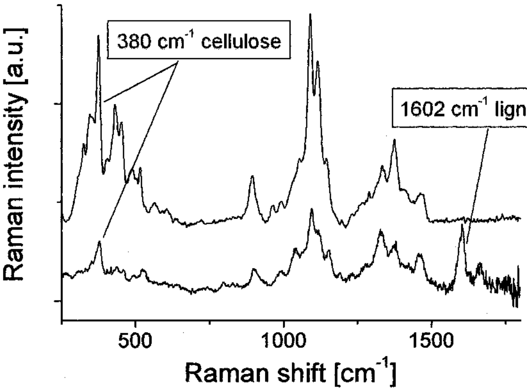

Figure 2. Raman spectra of beech wood (bottom) and of beech wood pulp (top).

The maximum lignin content was located in the corner of the cell as given in Figure 3 (b), whereas the cellulose content has a minimum there. The distribution of lignin and cellulose across a fibre cell wall is given in Figure 3 (a) representing the result of the measurements with an acquisition time of 30 s. The cellulose content decreased in the middle lamella and increased in the S2 layer. Additional information about the hemicelluloses content could not be gained, since the spectra of the hemicelluloses and cellulose cannot be separated as stated before.

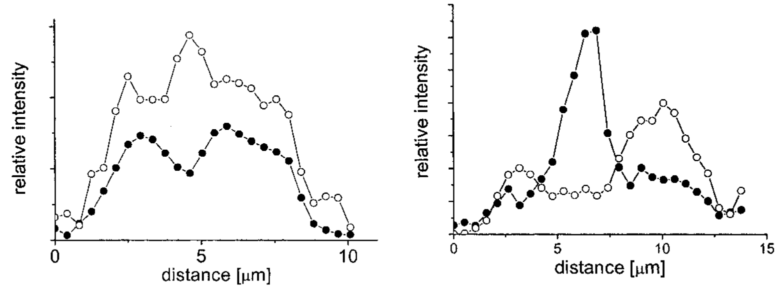

Figure 3. Distribution of lignin (—●—) and cellulose (—O—) in the cell wall (a) and in the cell corner (b).

These results verify the basic knowledge about the wood structure. The high spatial resolution of this method, however, allows the investigation of composites of wood and polymers for instance.

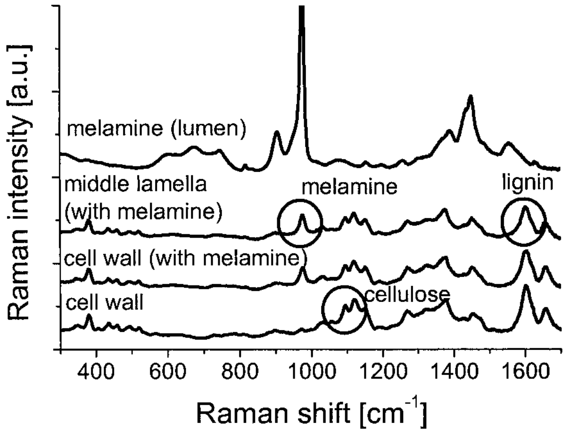

Figure 4. Raman spectra of melamine treated spruce wood in comparison to untreated sample (bottom).

Melamine-formaldehyde (MF) is one of the hardest and stiffest isotropic polymeric materials[9], which is used in decorative laminates, moulding compounds, adhesives, coatings and other products.

The lumen, which is filled with MF, was assumed to have a MF content of 100%. A sharp and intense band was observed at 975cm^{-1} (Figure 4, top), attributed to the triazin ring with a nitrogen radial in-phase vibration[10]. The intense ring vibration band at 974cm^{-1} was the only band recovered in the MF treated spruce cell walls (Figure 4) from the Raman spectrum of the pure MF resin. All other bands were also found in the untreated reference samples and can be assigned to cellulose, hemicelluloses and lignin[3]. A small band attributed to the cellulose at 965cm^{-1}was overlapped by the strong 974cm^{-1} melamine peak of the treated samples. The relative concentration was calculated by dividing the melamine peak area (993.9 cm^{-1} to 949.5cm^{-1}) of pure resin in the lumen with the peak area of the melamine within the cell wall. Figure 5 shows the calculated melamine content through a spruce wood cell. In addition the intensities of the cellulose (1079-1107cm^{-1}) and lignin bands (1568-1636cm^{-1}) in the Raman spectrum are lined out to verify the position within the cell wall (Figure 5). The average MF concentration detected within the cell wall of this section was 11.5%.

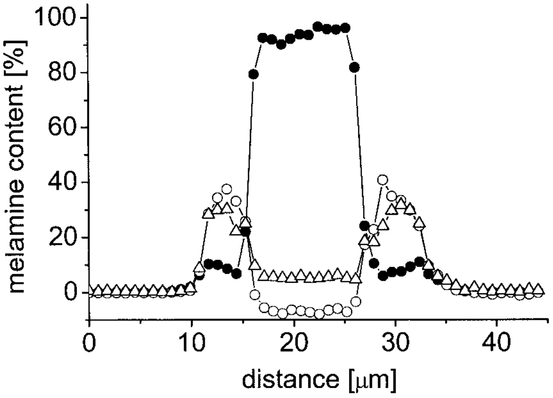

Figure 5. Distribution of melamine (—●—), cellulose (—△—), and lignin (—○—) in a spruce wood cell.

The cross sections of man-made cellulose fibres can be analysed by confocal Raman spectroscopy to get more information about their structure. The distribution of Raman active additives can be determined. Raman markers can be used to monitor the chemical and physical properties of the fibres, e.g. the usage of Calcofluor (Mw = 960 g/mol), a direct fluorescent dye. Lyocell fibres were treated various times with an aqueous solution of 1g/L technical Uvitex BHT (optical brightener, containing Calcofluor) and dried at room temperature. The distribution of this marker in the fibre can serve as a molecular probe for the structural properties of the fibre pore system

The intrusion of Calcofluor into the fibre cross-section is limited, depending on the time and the pore structure of the fibres. However, it can even reach the centre of the cross-section, especially by using never dried fibres with their open pore system, which decreases irreversibly after the first drying. The comparison between never dried and dried lyocell fibres measured by Raman spectroscopy is given in Figure 6. The Calcofluor content was determined in relation to the amount of cellulose, which was set as the internal reference.

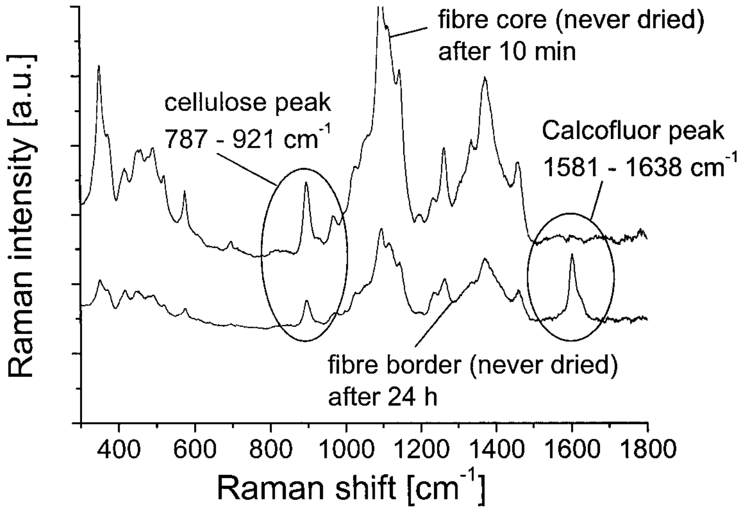

Figure 6a. Raman peaks of Calcofluor and cellulose used for the determination of Raman intensity ratio Calcofluor/cellulose in Figure 6b.

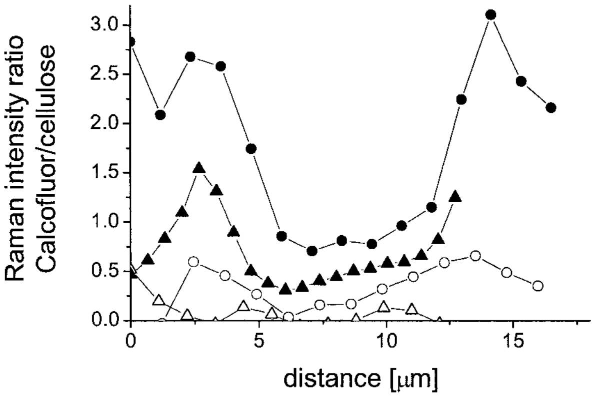

Figure 6b. Raman intensity ratio Calcofluor/cellulose (peak area 1581-1638 cm^{-1}/ peak area 787 – 921 cm^{-1}) for never dried, (—O—) after 1 h and (—●—) after 24 h, and dried lyocell fibre, (—△—) after 1h and (— —) after 24 h.

The investigation of the magnesium sulphite hydrate phase transition in a stirred vessel is an example for on-line Raman measurement. The spectra of the pure crystals of the two solid hydrates are given in Figure 7.

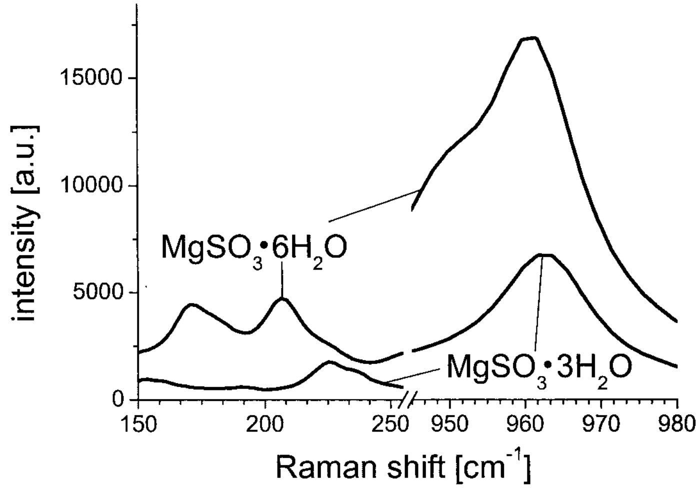

Figure 7. Raman spectra of solid magnesium sulphite trihydrate and hexahydrate

The different Raman signals of the hexahydrate and trihydrate allow the monitoring of the phase transition in real time. Therefore, the significant Raman shifts between magnesium sulphite hexa- and trihydrate bands were observed in the shift range of 150-250 cm^{-1} and from 950 to 980 cm^{-1}. The characteristical bands, used for the analyses were 172 cm^{-1}, 208 cm^{-1}, and 952 cm^{-1} for the hexahydrate, and 226 cm^{-1} and 962 cm^{-1} for the trihydrate.

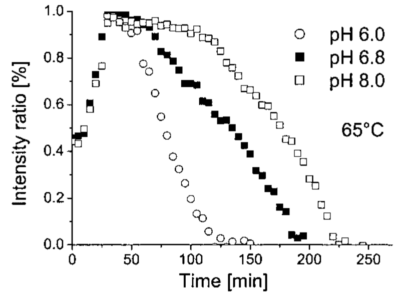

Figure 8a. Transition time of magnesium sulphite hexahydrate (65°C, pH range 6.0 to 8.0). Intensity ratio.

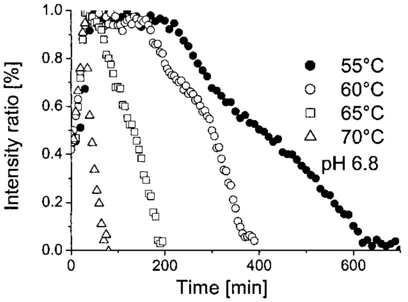

Figure 8b. Transition time of magnesium sulphite hexahydrate (pH 6.8, temperature range 55°C to 70°C).

The results show increasing conversion times with increasing pH-value at the beginning and with decreasing temperature. Therefore, an optimisation of the recovery process can be performed based on this information.

Conclusions

A growing field of application for the dispersive Raman spectroscopy can be provided by newly developed lasers, which can largely suppress the fluorescence with wavelengths close to the infrared combined with an higher spatial resolution compared to FT-Raman and IR spectroscopy. Inchoate from wood over pulp to regenerated cellulose fibres dispersive Raman spectroscopy can be used to determine content, modification, or structural properties in products along the "resource chain wood".

The lignin and cellulose distribution in the wood cell wall can be determined. The maximum content of lignin in the cell wall was found in the cell corners, whereas cellulose content reveals its minimum.

Melamine-formaldehyde (MF) and wood form stable composites, the distribution of the MF was determined with up to 11.5% in the wood cell wall.

The accessibility of regenerated cellulose fibres decreases during the drying process, which could be determined with Raman active marker compounds. The decrease in accessibility of lyocell fibres was monitored quantitatively during the drying procedure. Finally on-line measurements during phase transition of magnesium sulphite hexahydrate into trihydrate demonstrated an increasing reaction rate due to higher temperature and starting pH values, respectively.

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