Lignin−Quinone Interactions: Implications for Optical Properties of Lignin

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Charge transfer (CT) excited states are well-known and examined for artificial solution and solid-state donor−acceptor systems. Their possible existence in lignin has, however, not been considered before. In the present work we have studied the optical modifications resulting from the co-adsorption of the acceptor *p*-benzoquinone with selected donor compounds, some of which are related to lignin, onto filter paper. The quinone was also incorporated into the lignin of beech and spruce thermomechanical pulp fibers, and their optical absorption and emission properties were then examined. Evidence for the formation of CT states of donor compounds−quinone or (wood fiber) lignin−quinone is presented. Most notable is the formation of a broad-band visible absorption, at lower energies than what can be explained by quinone absorption alone, and the significant wood fiber emission quenching caused by these CT states. Since quinones commonly occur in various lignin types (e.g., wood pulps), this has the implication that lignin photo-oxidation may proceed via such CT states.

Introduction

The plant polymer lignin consists of various types of substructures covalently bonded into branched, disordered, polymeric network assemblies (Figure 1). The disorder is both chemical, with many different types of substructures, and conformational; that is, there is a distribution of relative orientations and separation distances between substructures. Thus, the electronic properties of lignin are considerably more difficult to predict from structure compared to those of artificial polymers, which usually contain only a single type of substructure. In this work, we deal with the lowest excited electronic singlet states of lignin, their excitation energies, and their absorption and emission properties—these states are responsible for the optical (UV−vis) absorption and fluorescence emission properties.

The plant cell wall emits fluorescence at wavelengths above ≈300 nm. This is commonly thought to be due to the aromatic lignin polymer.[1] Both lignin and the aromatic extractives (such as lignans, flavonoids, and tannins)[2−4] may have electronic excited singlet states of relatively low energy, and fluoresce at wavelengths above 300 nm, but in the current paper we will only refer to lignin. The interpretation of plant cell wall absorption and emission in the optical region (λ > 300 nm) includes therefore both lignin and extractives with lignin-like electronic properties as possible causes.

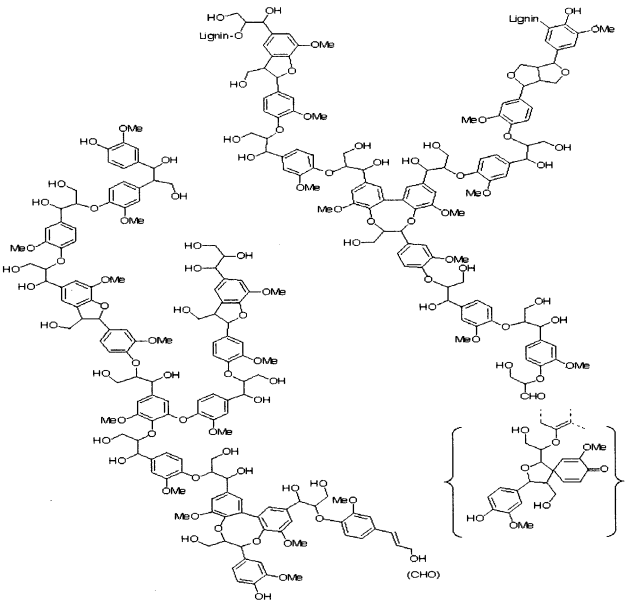

Figure 1. Tentative structure of lignin where typical substructure parts are indicated.

A description of the electronic states of lignin must take the properties of the individual substructure *n* as a starting point. A substructure consists of a minimum sized fraction of the lignin polymer, that is, a C_9 lignin monomer unit or—if a path of conjugation exists between two monomers—a lignin dimer unit (such as a stilbene-type unit). As a first approximation, lignin fluorescence of a sample *s* can be modeled as the total

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emission E arising from independent contributions, that is, a weighted sum of emissions $I(n, \upsilon_{em})$,

$$E(s,\upsilon_{ex},\upsilon_{em}) = \sum_n c(n,s)\, \epsilon(n,\upsilon_{ex})\, f(n,s)\, I(n,\upsilon_{em})$$

where $c(n,s)$ is the concentration, $\epsilon(n,\upsilon_{ex})$ the extinction coefficient, $f(n,s)$ the emission quantum yield, and $I(n,\upsilon_{em})$ the spectral shape of the emission of the substructure. Note that ϵ and I are sample independent basic properties of the substructure n, whereas the quantum yield f depends on the sample nature, that is, on the (average) molecular environment of n.

By the same principle the UV−vis absorption of lignin may be modeled by an absorption coefficient, which is a concentration weighted contribution of extinction coefficients, each of which is a fundamental property of a definite type of substructure.

This approach of relating optical properties with structural properties is based on the idea that the lignin polymer is a collection of *weakly* interacting chromophores (in a uniform dielectric medium) analogous to a dilute solution of a limited number of different types of chromophores, where the term "chromophore" denotes a set of electronic states with well-defined properties. Thus, each type of chromophore corresponds to a definite type of chemical substructure, an approach that has been used previously in the interpretation of plant fiber fluorescence.[1,4−7] It is, for example, a common approach to compare emission spectra of plant fiber or paper pulp samples to those of lignin model compounds. The model compounds are then assigned as the possible cause of the emission by comparing, for example, excitation and emission peak positions.

In the following we refer to this as *The Dilute System Assumption (DSA)*. This term is not commonly accepted but is adopted in the present work for the sake of clarity as well as for stressing the major point of this work.

For solution systems, the DSA may be invalidated by noncovalent intermolecular interactions. These may lead to the formation of excimers or exciplexes, where excited charge transfer (CT) states may be formed with properties that depend on those of the electron donor and acceptor compound, such as their oxidation and reduction potential, respectively.[8,9] Such CT states result in the formation of a chromophore, with characteristic absorption and emission, which *cannot* be described in terms of the donor or acceptor compound alone. Most notable is the creation of a CT absorption (or emission) band, which is considerably red-shifted relative to the lowest absorption band of the isolated donor or acceptor compound.

In a solid-state system, however, there are no free translations or rotations as in a solution system. These degrees of freedom are frozen (or relatively hindered), implying the possibility of stable time-invariant complexes of substructures, and therefore of stable time-invariant CT states, as is the case for crystalline CT materials consisting of co-crystallized donor- and acceptor compounds.[10−12] The lignin polymer is an amorphous solid-state system consisting of substructures that form time-invariant complexes. Within lignin we may find interunit separation distances as small as 4 Å,[13] which is a typical donor−acceptor distance for CT complexes. If two lignin substructures form a complex of favorable geometry, and have the properties required for formation of a low-energy excited CT state, that is, a low oxidation potential and a high reduction potential, respectively, then the optical properties of lignin may contain contributions from such states.

The basic elements $\{n\}$ of the DSA, lignin chromophores, may not only be lignin monomer units, or certain dimer units, but may also consist of larger size systems, such as lignin CT donor and acceptor substructure complexes. Thus, for example, photo-oxidation (photoyellowing) of paper may also proceed via lignin low-energy CT states, which are formed through interactions between at least two substructures. In this case, the idea of a single type of substructure responsible for photoyellowing does not apply.

In the present work we describe the experimental formation of lignin-based, low-energy, excited CT states in plant cell walls and their effect on the optical properties of lignin. The electron acceptor, *p*-benzoquinone, is shown to form excited CT states with selected electron donor compounds—some of which models lignin substructures—when coadsorbed on filter paper. *p*-Benzoquinone is also added to hardwood and softwood fibers and allowed to diffuse spontaneously into the cell wall. The interaction of the lignin with this quinone and the resulting change of material properties is analogous to the molecular doping of conjugated polymers.[14] This quinone bears structural resemblance to oxidation products derived from lignin substructures. Quinones are also known to be good electron acceptors in charge transfer (CT) excited states of donor−acceptor complexes in solution.[8,9] The effect of the quinone on the optical properties of the fibers is examined, and evidence for the formation of lignin CT states due to the presence of the quinone is presented.

Materials and Methods

Wood Fibers and Paper. *Hardwood*. Beech (*Fagus sylvatica*) thermomechanical pulp (TMP) fibers were supplied by the MDF (medium density fiberboard) plant of Junckers Industries, Køge, Denmark, where the fibration is carried out using an Asplund process. The fibers were frozen after fibration, thawed, and conditioned to the ambient environment just before usage.

Softwood. Norway spruce (*Picea abies*) TMP fibers were supplied by Sunds Defibrator, Sundsvall, Sweden, where the fibration is also carried out using an Asplund process, and handled as the beech fibers before usage. Filter paper for droplet treatments was 589[1] Black ribbon from Schleicher & Schuell.

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Chemicals. 1,4-Dioxane was obtained from Fisher Scientific and ethyl acetate from Bie & Berntsen A/S. *p*-Benzoquinone, catechol (1,2-benzenediol), resorcinol (1,3-benzenediol), and vanillyl alcohol (4-hydroxy-3-methoxybenzyl alcohol) were obtained from Sigma-Aldrich and crystallized by sublimation. The lignin monomers coniferyl alcohol and sinapyl alcohol were prepared by the method of Lu and Ralph.[15] The benzenediols, vanillyl alcohol, and lignin monomers are referred to as "donor compounds".

Droplet Deposition Treatments. An ethyl acetate solution containing 0.1 M purified quinone and 0.1 M donor compound was prepared. A 100-μL droplet of the solution was added to a sample of filter paper kept under argon or air. The ethyl acetate was allowed to evaporate and the sample transferred to an UV−vis spectrometer to monitor the optical modification initiated by the solvent evaporation.

Fiber Treatments. Five grams of dry matter of each fiber type was first subjected to a Soxhlet hot de-ionized water extraction for 24 h[16] and then dried for 24 h at 40 °C. This minimizes the possibility that the quinone should subsequently alter fiber/solution sorption equilibria of water-soluble extractives. Such an effect could interfere with that caused by the quinone.

Sorption of the quinone into the fibers was done by adding 1% (by weight) of dry fibers to an aqueous solution of the crude quinone at 20 °C. Quinone solutions were prepared in concentrations {0, 0.10, 0.010, 0.0014 M} for beech and {0, 0.10, 0.011, 0.0012 M} for spruce fibers. The fibers buffered the solutions to ca. pH 5. The suspensions were then left for 24 h at 20 °C after which the solution was removed and the fibers washed thoroughly with de-ionized water and dried as above. The treatment takes advantage of a sorption nonequilibrium for the quinone in relation to the fiber/water phases.[17] Hence, the fibers contain adsorbed quinone, except the control fibers prepared without the quinone.

The two treatments with maximum quinone concentration, and the two controls, were repeated with (1) nitrogen flushing of the solution during the 24-h treatment time and (2) with replacement of the crude quinone with the purer crystallized quinone.

Extraction of *p*-Benzoquinone from Fibers and Detection by Gas Chromatography/Mass Spectrometry (GC-MS). To verify the presence of *p*-benzoquinone, fibers were extracted with dioxane and the extract was analyzed. The fibers were suspended at 5% consistency in a solution of 90% dioxane and 10% water. Extraction was done for 24 h under argon flushing. Then 10 mL of the extract was filtered and frozen immediately after extraction. Samples were prepared by evaporating the extracts at 40 °C under nitrogen flushing after which they were redissolved in 0.5 mL of dioxane.

GC-MS of the concentrated extracts was done by a Perkin-Elmer Autosystem GC and Turbomass MS detector. The column was a CPSIL 19CB o.d. 0.32-mm column, total length 60 m. Carrier gas was helium with a flow rate of 1 mL/min. Injection volume 1 μL. Temperature profile: initial temp/hold 55 °C/2 min, ramp at 20 °C/min to 240 °C, hold for 2.75 min. Total run time was 14 min.

Spectroscopy. Paper samples were analyzed by a Cintra 40 UV−vis spectrometer equipped with an integrating sphere, using a barium sulfate disk as a 100% reflectance standard.

Fiber sample disks of 1-mm thickness were produced by a Perkin-Elmer hydraulic press (at 5 bar). Instruments used were a Perkin-Elmer Lambda 9 UV−vis−NIR spectrometer, using a barium sulfate disk as a 100% reflectance standard, a Perkin-Elmer 2000 FT-IR, and a SPEX 1680 0.22-m double-fluorescence spectrometer in front-face setup.[18] For each sample type five independent measurements were made.

For Raman spectroscopy and XPS (X-ray photoelectron spectroscopy) only beech fibers, 0.1 M quinone and control, were measured. Samples were ball-milled for 1 h using a Retsch MM2000. The container was regularly cooled to minimize sample degradation.

Raman measurements were done using an IFS 66 FT-IR instrument equipped with a Bruker FRA 106 FT-Raman module. A 300-mW Nd:YAG laser of 1064 nm was used in a 180° scattering configuration, and the detector was a Ge-diode cooled to liquid nitrogen temperature. Each spectrum was obtained as the average of 1000 scans with a spectral resolution of 6 cm^{-1} (after apodization). Three independent measurements (spectra) were made for each sample type.

XPS measurements were done with a Specs Sage 100 instrument. A sample was prepared by placing the powder material onto double-sided tape in a thick layer such that no signal arose from the tape itself. Three independent spots from each sample type were analyzed. Elements were identified and quantified from survey spectra (0−1100 eV), and curve fitting was peformed on the high-resolution carbon C 1s and oxygen O 1s spectra.

All measurements, except XPS (which is done in a vacuum), were done under ambient conditions.

Results

The paper samples obtained from droplet deposition treatments were analyzed a few minutes after the deposition. For droplets containing both quinone as well as donor compound the samples were initially pale yellow as the sample was still wet. When the solution evaporated after ≈1 min, a new color (red-to-brown depending on donor type) formed at the edge, which within a few seconds spread to the entire area of the droplet. In Figure 2 representative remission functions showing this phenomenon is depicted. All functions are depicted *relative* to pure filter paper, that is, as the difference to the average of five independent filter paper UV−vis remission functions. These filter paper remission functions were identical (within $\Delta F = 0.001$ or $\Delta A = 0.002$) over the whole wavelength range. When the samples were extracted with ethanol, the new color disappeared from the samples and was *not* present in the extract, which reflected the characteristic UV−vis absorptions of the quinone and the donor, respectively (results not shown). When samples were kept under ambient conditions, the newly formed color disappeared (in a time scale of a few hours), leaving back a pale yellow color. Deposition of droplets containing only the quinone or only a donor compound resulted in an unaltered color with no color transition. For the quinone the yellow color faded away, that is, the absorption band

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(16) One may object to the severity of such a treatment, which subject the fibers to both physical and chemical changes. But first, it can be noted that the process producing such fibers (i.e., the refining) is itself a severe treatment; second, we are *not* interested in a characterization of the TMP fibers as such, but in a characterization of a chemical/physical *difference* which is induced by the presence of a quinone.

(17) Initially, we attempted the alternative method (used in the preparation of two-component crystals, see: Ito, Y. *Synth.-Stuttgart* **1998**, *1*, 1) of simply mixing the quinone (powder) with the dry fibers in a ball mill hollow cylinder (25 mL) in which a stainless steel ball (7 g) provides mechanical and thermal energy when the cylinder is shaken (by a Retsch, MM2000). We did, however, find that this method invariably produced significant chemical transformations of the quinone (as detected by UV−vis spectroscopy of the solution obtained by adding deionized water to the resulting sample) due to the mechanical energy. Therefore, this method was discarded. The method used in the current work is analogous to a sublimation of the quinone, which sublimes *within* the wood cell wall.

(18) Due to the high light-scattering ability of plant fiber materials, it is important to use a fluorescence spectrometer with at least two monochromators at each side (excitation and emission) as the one used for this work. By the use of a nonfluorescent highly scattering powder (e.g., barium sulfide), it is easily seen that single monochromator instruments give rise to spurious effects.

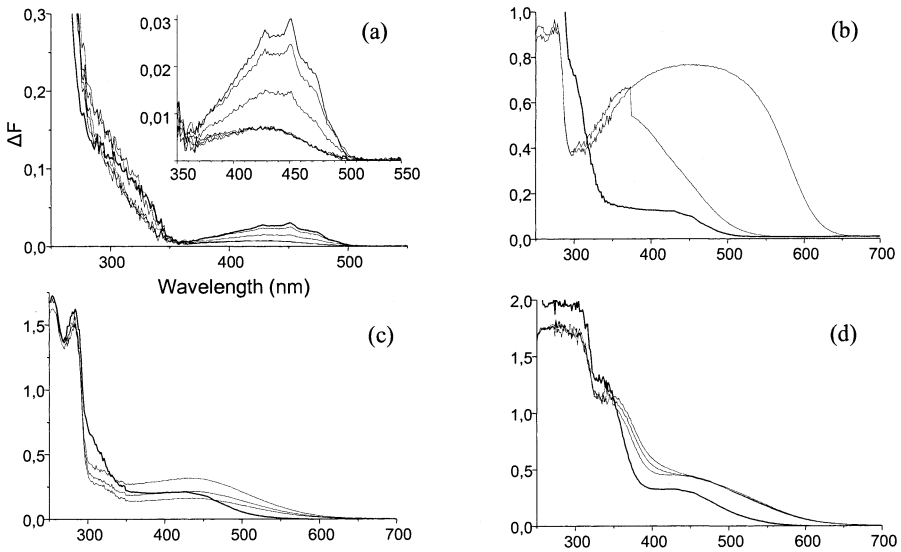

Figure 2. (a) UV−vis difference remission functions of *p*-benzoquinone adsorbed on filter paper, 5 min (top spectrum) and 10, 15, 20, 30, and 40 min (bottom) after droplet deposition. (b) Recorcinol and *p*-benzoquinone co-adsorbed. In order at 400 nm: 5 min (bottom), 10 min (top), and 15 min (middle) after droplet deposition (the small discontinuity at 375 nm of one function is an instrumental artifact). (c) Coniferyl alcohol and *p*-benzoquinone co-adsorbed. In order at 500 nm: 2 min (bottom) and 5, 10, and 15 min (top) after droplet deposition. (d) Vanillyl alcohol and *p*-benzoquinone co-adsorbed. In order at 400 nm: 2 min (bottom) and 5, 10, and 15 min (top) after droplet deposition. The units are Kubelka-Munk units $F(R) = (1 - R)^2/2R$. The remission function drawn with a thick line is the initial remission function, that is, after 2 min for (c) and (d) and after 5 min for (a) and (b).

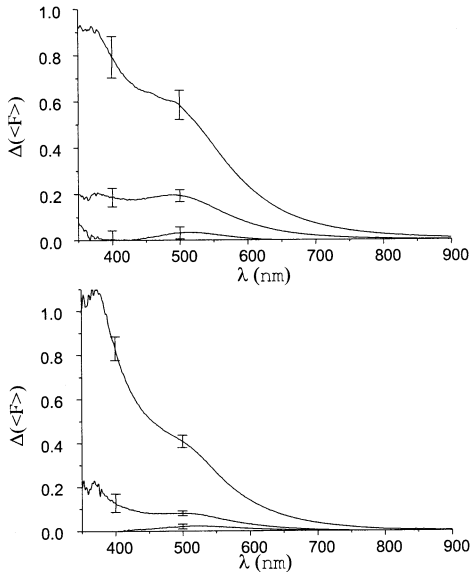

Figure 3. UV−vis difference remission functions of quinone-treated samples (top, beech; bottom, spruce). In this figure, as well as in the following, error bars are calculated from standard deviations of the mean value.

at 440 nm decreased with time and its initial vibronic structure disappeared; see Figure 2.

For wood fibers the UV−vis remission function *relative* to the control sample is depicted in Figure 3 for each quinone-containing fiber sample. The effect of the presence of the quinone in the fibers is significant. For both softwood and hardwood fibers, the quinone causes two main absorption bands: A high-energy band with an onset located at ≈400 nm and a broad, structureless, low-energy band with maximum absorption around 500 nm, but with an onset located at much smaller energy. With increasing amount of adsorbed quinone the intensity of these bands increases with a relatively higher increase of the high-energy band. It is noteworthy that these qualitative features are invariant with respect to

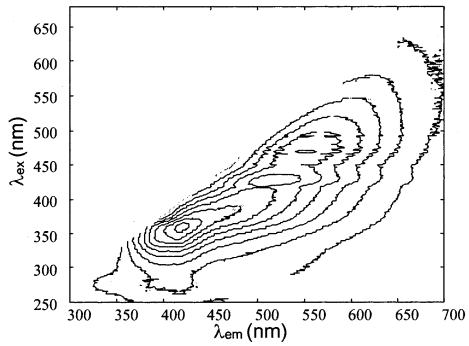

Figure 4. 2D contour plot of the emission intensity of a beech control sample as a function of excitation and emission wavelength.

the two different wood species. At wavelengths below 400 nm the instrumental noise becomes too large to merit any conclusive observations.

For both beech and spruce control fibers the low-energy excitation range (>400 nm) shows a broad fluorescence emission peak, which is always red shifted by approximately 100 nm from the excitation wavelength. This is exemplified for beech control in Figure 4.

The effect on the fiber fluorescence due to the presence of the quinone is pronounced. A significant quenching (reduction) of the emission is observed. This is shown in Figure 5 for the beech samples. Quenching is observed for all excitation wavelengths, and the degree of quenching correlates with the quinone concentration applied in the sorption treatment. Again, these results are invariant with respect to the two different wood species.

In Figure 6 is shown the unitless factor α, which is a function of the excitation wavelength λ_{ex} and the degree of quinone sorption, by which the control emission spectrum $I_C(\lambda_{em})$ is multiplied in order to approximately obtain the emission spectrum $I_D(\lambda_{em})$ of a quinone-

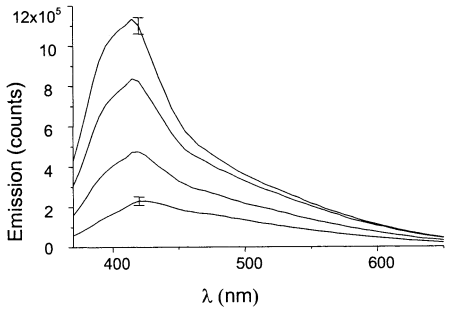

Figure 5. The most prominent emission spectrum of the beech samples (λ_{ex} = 350 nm). The top curve is the control. The three lower intensity emissions are the quinone-treatment samples with the lower the emission, the higher the quinone concentration of the doping treatment.

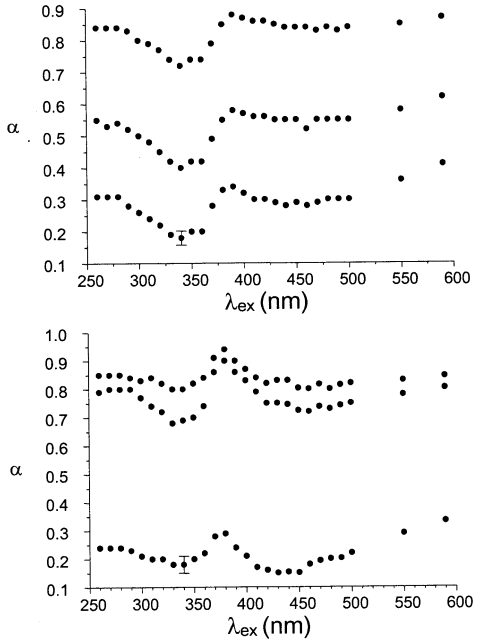

Figure 6. Quenching factor as a function of excitation wavelength for each quinone-treated treatment (control treatment has factor of 1 by definition). Top, beech fibers; bottom, spruce fibers.

treated sample, that is, $I_D(\lambda_{em}) \approx \alpha I_C(\lambda_{em})$. For the derivation of this quenching factor the wavelength of maximum emission, for each excitation wavelength of the control fibers, was chosen. Thus, full emission quenching corresponds to a factor of 0, whereas no quenching, or unaltered emission, corresponds to a factor of 1.

A clear excitation wavelength dependency is seen with maximal quenching at 350 nm for both spruce and beech. But significant quenching is observed even at the lowest excitation energies corresponding to emission energies *well* below the absorption edge of the (in solution) weak n → π^* quinone transition. The functional shape of $\alpha(\lambda_{ex})$ is roughly invariant with respect to the quinone concentration and fiber type. The fact that the beech and spruce fibers with lowest quinone concentration have a negligible UV−vis excess absorption is not reflected in the quenching of their emission, which is significant. All the observed spectral modifications are thus qualitatively identical for both spruce and beech fibers.

In the NIR−IR/Raman spectral region no effect of the quinone could be detected, as the spectra of the control fibers are indistinguishable from those of quinone-treated fibers. Likewise, the XPS measurements revealed no clear differences between quinone-treated fibers and control fibers.

The GC-MS measurements reveal the presence of the quinone in the quinone-treated fibers as a signal peak, which increased with increasing quinone concentration, the identity of which is clearly established by MS (results not shown). In addition, another small peak, of molecular weight 73 Da, was seen in the quinone-treated fibers, but not in the control. Hydroquinone was not observed, nor was any other compound of high molecular weight (>108 Da) observed to differentiate the quinone-treated from the control fibers.

No differences were found between fibers treated under anaerobic ambient conditions and fibers treated also under nitrogen flushing, nor had application of a more purified quinone in the sorption treatment any influence on the results.

Discussion

UV−Vis Spectroscopy. The droplet deposition experiments clearly show that when the quinone acceptor and one of the donor compounds are adsorbed together, new CT bands appear, which are not just independent contributions from either compound. Subsequent extraction with ethanol demonstrated the reversibility of their formation.

This requires a sufficiently large (ground state) population of the donor−acceptor complex relative to the population of noninteracting donors or acceptors. The strength of the donor−acceptor ground-state interaction—hence the size of the population of the complex—is primarily determined by the difference between the ionization potential of the donor and the electron affinity of the acceptor. It is well-known that, in order for the CT state of the complex to be observable in the optical energy range, that is, optical states, this difference must be relatively small such that the CT state has low energy. In addition to this the geometry of the complex is also important.

The fact that the quinone−donor compound solutions did not form observable CT bands *in* solution but first on a paper sample upon solvent evaporation demonstrates the first (thermodynamic) requirement. The ground-state interaction between donor and acceptor in solution cannot compete with their respective interaction with the solvent. When, however, the solution is replaced with the paper surface, the donor−acceptor interaction becomes relatively stronger, ensuring a sufficient population of complexes.

It is, however, only a quasi-equilibrium since the population of complexes eventually decreases. The behavior of the remission function of the sample with quinone deposited (alone) shows an absorption decrease of the low-energy n → π^* transition (at 440 nm), which is *not* accompanied by a decrease of the $\pi \rightarrow \pi^*$ transition (at 290 nm). Hence, this does not show the evaporation of quinone from the sample, but rather the fact that hydrogen bonds between cellulose hydroxyl groups and the quinone is formed. This interaction may cause the initially formed complexes to dissociate as it eventually competes with the interactions leading to complex formation with donor compounds.

The requirements for the formation of optical CT states with the quinone were evidently fulfilled, even for vanillyl alcohol as the donor. Since this compound is a good model for a majority of lignin substructures, and since some of these have even better donor properties (lower ionization potentials), this observation of CT state formation with the quinone offers further support to the general notion of quinone−lignin CT states.

For fibers treated with quinone, the excess UV−vis remission function does not reveal the characteristic n $\rightarrow \pi^*$ quinone transition (aqueous solution, 430 nm; filter paper, 440 nm). It reveals on the other hand a low energy band, which does *not* have any corresponding transition of the quinone in solution, or on filter paper, to which it can be assigned. For the highest quinone concentrations, it clearly extends beyond a wavelength range, which can be explained by absorption of a noninteracting quinone (compare Figure 3 with Figure 2a).

In principle, a large variation with wavelength (over the range 400−600 nm) of the fiber scattering properties could enhance the low-energy tail of the n $\rightarrow \pi^*$ quinone transition, giving rise to an apparent new transition band since the Kubelka−Munk function F is proportional to K/S, where K is the (fiber) absorption coefficient and S the scattering coefficient.[19] This would require the scattering properties to be significantly modified due to the quinone treatment. Such modification would imply significant changes in particle size, which is only accomplished by gross removal and degradation of cell wall material. The results of the IR and Raman measurements show that this is definitely not the case.

This band is thus caused by the presence of the quinone, but in a way which implies electronic interaction with other molecular substructures in the fibers. In accordance with the current knowledge of CT states formed between donor−acceptor compounds in solution, and in accordance with the results of the droplet deposition experiments, these observations show the formation of low-energy lignin−quinone CT states.

Fluorescence Spectroscopy. For all fiber samples the low-energy excitation range ($\lambda_{ex} > 400$ nm) shows a broad fluorescence emission peak, which is always red-shifted by ≈100 nm from the excitation wavelength. This indicates a band structure (i.e., continuous distribution) of the excited electronic states of the lignin. This is likely true of the whole UV−vis excitation energy range due to the chemical and physical heterogeneity of lignin. Hence, any attempt to isolate emission components will likely isolate contributions corresponding to sub-bands caused by a variety of different types of substructures. Therefore, the notion of a Stokes shift is strictly speaking not meaningful when applied to the observed excitation−emission peak energy shift.

The results of the fluorescence measurements demonstrate the occurrence of excitation energy transfer (EET) in lignin.[20−22] The samples compared are fiber lignin without quinone and fiber lignin with quinone incorporated, in other words, a population of emitting substructures *with* and *without* the nonfluorescent quinone. Since none of the emitting substructures disappear (or are transformed) due to the sorption treatment, they pass on their excitation energy (and become nonemitting) by EET to lignin−quinone CT states, which are evidently nonemissive. This demonstrates electronic interaction between lignin band states, leading to EET, which depends sensitively on the relative orientation and distance between substructures such that changes in the conformational (physical) state of lignin may result in the creation or destruction of apparent emission components.

It may be argued that the emission quenching is only an effect of re-absorption by the quinone; that is, it absorbs the fluorescence emission. This suggestion is, however, difficult to reconcile with an up to a factor of 6 reduction of fluorescence at 400 nm compared with the relatively more modest increase of absorption around 400 nm (see Figure 3), as well as with the fact that even the samples with the lowest quinone concentrations show significant quenching despite a negligible excess UV−vis absorption.

The fact that the quenching factor $\alpha(\lambda_{ex})$ varies with excitation energy adds another reason (apart from the band nature argument) why the assignment of an emission peak to a definite type of chemical substructure should be approached with caution. Lignin is known to contain quinones produced by oxidation or photo-oxidation.[23−25] As demonstrated in this work, these may quench the emission via EET from other compounds, the emission of which may then not contribute much to the overall fluorescence. Even for a single type of quinone, the emission from other compounds will be unevenly quenched as demonstrated by $\alpha(\lambda_{ex})$. According to the expression $E(s,\upsilon_{ex},\upsilon_{em})$ for the total emission (see the Introduction), this effect is incorporated into the individual substructure quantum yield $f(n)$. This of course means that no simple relationship exists between (1) the concentration of a substructure in lignin and its quantum yield $f_{sol}(n)$ in solution (where $f(n) \neq f_{sol}(n)$) and (2) its relative contribution to the total emission. This expresses the fact that decay channels of lignin excited states include an excited-state interaction such as EET.

GC-MS and Raman Spectroscopy. The Raman and GC-MS measurements in combination show that the sorption treatment results in the physical incorporation of the quinone in the fiber material (GC-MS), but that the amount of quinone in the fibers is small (Raman). This is stressed by the fact that the strong *p*-benzoquinone Raman transitions at 1600−1700 cm^{-1} [26,27] cannot be detected, even in the fibers containing most quinone. Furthermore, the XPS results show that the quinone cannot be detected.

The extra GC-MS peak at 73 Da found in the quinone-treated fibers is believed to be caused by changing solubility constants of extractives/volatiles caused by the

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presence of the quinone in the cell wall, and not by reactions of the quinone with cell wall components; that is, the quinone is assumed not to be chemically reactive under the applied conditions. This assumption is supported by the fact that UV−vis results of the droplet deposition experiments were reversible, that is, the observed color transitions were not due to, for example, irreversible addition reactions. For the fiber samples such reactions should yield products observable by GC-MS. But this was not the case.

Perspectives. These results are similar to those obtained in a previous work where an enzyme (laccase) catalyzed modification of the same type of spruce and beech fibers was examined.[28] It was suggested that the laccase treatment produced lignin radicals, some of which decayed into oxidation products, and that these products were indirectly observable due to the fact that they quenched the fiber fluorescence.

In the present work we have added to the feasibility of this suggestion since the quinone is a reasonable model of a lignin oxidation product. The degree of quenching increases with the amount of added quinone, analogous to the fact that for the enzyme-treated fibers the degree of quenching increased with the amount of enzyme added (i.e., the oxidative capability of the treatment). For the enzyme-treated fibers the oxidation products could not be detected by other means (i.e., IR spectroscopy), which indicated that they occurred in low concentration, and in the present work only GC-MS afforded a direct detection of the quinone. This implies that fluorescence properties are extremely sensitive to the occurrence of lignin oxidation products, that is, the occurrence of nonemissive low-energy excited states. This is well-known for materials of conjugated polymers, where carbonyl defects have a large impact on the polymer emission, which decreases significantly, even for very small defect concentrations.[14,29]

The realization that CT states have a considerable impact on the electronic properties of lignin is relevant for the understanding of photo-yellowing of pulp and paper. In research within this field efforts are often directed toward finding and characterizing chromophores responsible for photo-yellowing. The present work has indicated that a chromophore, such as *p*-benzoquinone, may gain entirely new electronic properties upon transfer from the liquid phase to solid-state lignin. Hence, its response toward photons, which depends on its electronic state nature, differs entirely in the solid state than in solution, and so do (photo-)chemical reaction pathways from its excited states too. Furthermore, it has been demonstrated that EET is a phenomenon, which does occur not only in conjugated polymers or doped molecular crystals[14,29−31] but also in lignin where nonemitting CT states may accept excitation energy from other lignin chromophores. These aspects could therefore be useful to consider in the photo-yellowing research on plant fiber material.[32]

Conclusion

The identification of lignin chromophores, or sets of electronic states, which are elements in the DSA, with definite chemical substructures is not trivial. Low-energy excited CT states can be formed due to donor−acceptor interactions between substructures, which has been clearly demonstrated in the present work by incorporating *p*-benzoquinone into wood fiber material. Such states exemplify chromophores consisting of several substructures, and in the disordered lignin polymer a wide distribution of CT state properties is to be expected, reflecting chemical (oxidation and reduction potentials) as well as structural (interunit separation distance and orientation) disorder. They occur in both softwood as well as hardwood lignin and are thus a generic property of lignin. CT states have a large impact on the absorption and emission of lignin. Their impact on lignin fluorescence properties demonstrate that excited-state interactions between the elements of the DSA, that is, between lignin chromophores, play an important role in the photochemical reactions of lignin.

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