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NEW INSIGHTS INTO CHROMOPHORE CHEMISTRY OF EUCALYPT PULPS ASSESSED BY UV-RESONANCE RAMAN MICRO-SPECTROSCOPY

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SUMMARY

UV-Resonance Raman (UV-RR) spectroscopy with laser beam excitation source of 325 nm was applied to a solid-state study of chromophores in *Eucalyptus globulus* kraft pulps. The amount of chromophore structures in pre-bleached/fully bleached pulps absorbing at 325 nm linearly correlated with signal at ca 1600 cm^{-1} in UV-RR spectra. The characteristic vibrations from particular pulp chromophore structures have been assigned from experiments with model compounds thus allowing establishing an UV-RR database. Among the components of bleached eucalypt kraft pulp, xylan-lignin complex was suggested to be an important source of chromophores. The monitoring of pulp bleaching in ECF sequences by UV-RR allowed a proposition about formation of new polysaccharide-derived chromophores that hindered the brightness development in the final D stages. These chromophore systems are co-responsible for the brightness reversion of fully bleached pulps during thermal accelerated ageing and are vulnerable to alkali extraction stages in standard ECF sequences. This study revealed UV-RR spectroscopy as a promising tool for quantitative monitoring of chromophores during pulp bleaching and its relationship to thermal ageing.

Keywords: Ageing, Bleaching, Chromophores, Eucalyptus kraft pulp, Raman spectroscopy

INTRODUCTION

Bleaching operations contribute significantly to the cost of chemical pulps and are far of technical perfection. In effect, the last bleaching stages, which are projected towards attaining high ISO brightness of pulps (≥ 90 %), are of particularly low efficiency regarding the brightness development. Further improvement of bleaching technologies implies a better understanding of the origin and reactivity of chromophore structures in pulps. This knowledge would reveal the relation of chromophores to brightness gain and brightness stability of the fully bleached pulps.

The chemistry of chromophores is one of the less investigated topics in the area. This lacuna is mostly due to difficulties in assessing such a minor amount of chromophore structures in semi-bleached and bleached pulps (level of ppm or even ppb) which instability hinders their concentration and unaltered isolation. In fact, the origin and structure of chromophores are mostly speculated though this information is essential to understand the oxidation mechanisms of cellulose, hemicelluloses and residual lignin. Therefore special attention is paid to advanced analytic techniques allowing non-invasive chromophore detection and identification. In this context, Raman spectroscopy presents a unique tool towards a highly sensitive *in-situ* solid-state assessment of minor moieties in cellulosic pulps. An important feature of Raman spectroscopy relies on using enhancement techniques for the Raman scattering signal, such as Resonance Raman Spectroscopy (RRS) or Surface Enhanced Raman Spectroscopy (SERS). The RRS allows a selective signal enhancement of 10^2-10^6 times whenever the excitation wavelength is close to an electronic absorption peak. For that reason, UV excitation is well

suited to selectively record the Raman spectra of absorbing structures belonging to complex and heterogeneous biologic systems such as cellulosic pulps. Additionally, the use of UV excitation allows further overcoming of strong laser-induced fluorescence since this type of luminescence becomes farther from the Raman region, thus avoiding Raman and fluorescence bands overlap.

The potential of Ultraviolet-Resonance Raman (UV-RR) spectroscopy for the analysis of residual lignin and hexeneuronic acids (HexA) in chemical pulps have been demonstrated in previous studies using excitation at 244 and 257 nm, close to the absorption maximums of those structures [1, 2]. However, HexA and most part of lignin do not contribute to the colour of pulp, while colour-inducing structures (conjugated carbonyl and polyunsaturated structures among others) possess the maximum absorption in the range of 300-400 nm. A selective detection of highly conjugated chromophore structures in bleached kraft pulps is possible using 325 nm laser beam excitation (@ 325 nm) , thus avoiding the contribution of HexA and unmodified lignin to specific chromophore bands in UV-RR spectra [3]. The proposed approach allowed the monitoring of chromophores in pulps at ca. 1600 cm^{-1} and to track the extent of polysaccharide oxidation upon bleaching at 1093 cm^{-1}. The existence of charge transfer complexes (CTCs) in pulps that combine chromophores and oxidized moieties in carbohydrates were suggested. These CTC can affect the intensity of the aforementioned bands by diminishing the conjugate state in the chromophore moieties thus limiting the quantitative analysis of Raman spectra [3].

This work deals with further developments on application of UV-RR spectroscopy for the study of chromophores in cellulosic pulps. UV-RR was coupled to UV-visible diffuse reflectance (UV-vis DR) spectroscopy to provide a fundamental background for the assignment of characteristic vibrations of particular chromophore structures employing a series of model compounds. This knowledge was expanded for the interpretation of chromophore changes in industrial *Eucalyptus globulus* kraft pulps along two different ECF bleaching sequences (DEDED and OQ(PO)DP). Additionally, bleached pulps submitted to wet-thermal ageing were analysed employing UV-RR spectroscopy and the origin of chromophore structures responsible for the brightness reversion has been proposed.

EXPERIMENTAL

Materials

The industrial unbleached and ECF bleached *Eucalytpus globulus* kraft pulps (DEDED and OQ(PO)DP of 91.5 ± 0.5% ISO brightness) were supplied from two Portuguese pulp mills. The pulps were further washed in the laboratory with distilled water and conditioned in a dark at +4 ºC. Another set of industrial pulps, bleached by DEDED or DEDD sequences (ISO brightness range of 89–91%) were subjected to wet-thermal ageing.

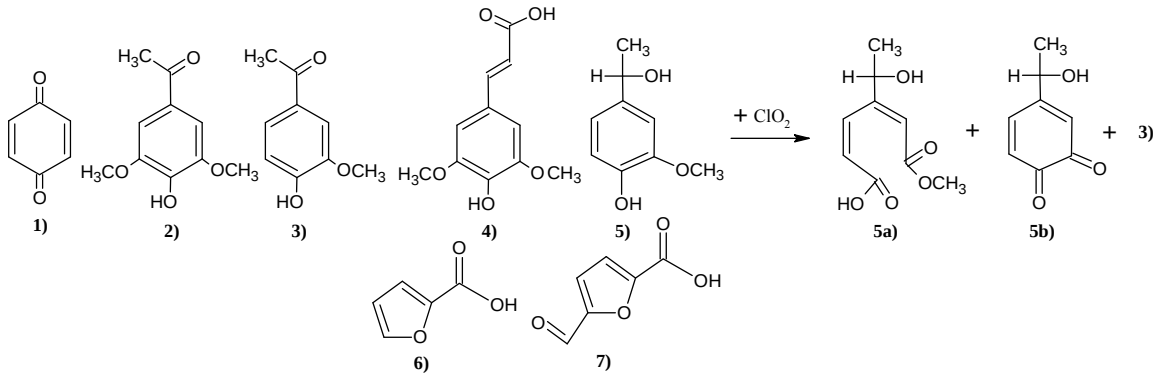
Cotton linters and model compounds (Fig. 1) of p.a. grade were supplied by Aldrich Chem. Comp.: **1.** *p*-benzoquinone; **2.** 3,5-dimethoxy-4-hydroxyacetophenon; **3.** 4-hydroxy-3-methoxyacetophenon (acetovanillone); **4.** 3,5-dimethoxy-4-hydroxycinnamic acid; **5.** 4-hydroxy-3-methoxy-α-methylbenzyl alcohol (apocynol); **6.** 2-furancarboxylic acid (2-furoic acid); **7.** 5-formyl-2-furancarboxylic acid. Compounds **5a** (muconic acid derivative) and **5b** (o-quinone derivative) were obtained by oxidation of **5** with chlorine dioxide according to the procedure described previously [3]. Typically 3 mg of each chromophore model was dissolved in 1 ml of acetone and mixed with 100 mg of cotton linters. Eucalypt kraft lignin, precipitated from black liquor, was dissolved in dioxane and mixed with cotton using similar proportion as with model compounds. Acetone and dioxane were evaporated in N_2 stream at room temperature under dark conditions. All cotton samples with deposited model compounds were pressed into 11 mm diameter pellets during 30 s at 50 MPa.

Birch (*Betula pendula*) xylan was isolated by extraction of peracetic holocellulose with 5% KOH. HexA-enriched xylan was obtained by treatment of xylan in 1M NaOH solution at 150 ºC during 30 min. Xylan samples were analysed as 3 mm diameter pressed pellets.

Methods

Pulp handsheets were prepared according to ISO 3688. The measurement of ISO brightness (ISO

2470) was carried out using L&W Elrepho SE 070 spectrophotometer. Brightness reversion was performed under wet-thermal conditions (100 % R.H. and 100 ºC) according to TAPPI T 260.



.**Figure 1. Model compounds mimicking chromophores and degradation products of cellulosic pulps.**

UV-vis Diffuse reflectance (UV-vis DR) spectra of 11 mm diameter pellets (e.g., cotton with deposited model compounds) and round-cut pulp handsheets (the same used for ISO brightness measurements) were recorded on a JASCO V-560 spectrophotometer equipped with a JASCO ISV-469 integrating sphere and using $BaSO_4$ standard as background reference. The range was 200-800 nm with a scanning speed of 200 nm/min and a bandwidth of 5 nm. The reflectance spectra were also converted into k/s spectra using known Kubelka-Munk Eq. (1):

$$\frac{k}{s} = \frac{(1-R)^2}{2R} \quad (1)$$

where R is the reflectance of the opaque sample, k is the specific absorption coefficient and s is the specific scattering coefficient (s was the same for all samples).

Micro-Raman spectra of pellets were recorded on a Jobin Yvon (Horiba) LabRam HR 800 micro-Raman spectrometer @ 325 nm (He-Cd UV laser, Kimmon IK Series) under backscattering configuration and using a 40X NUV objective. The spectral range was 750 – 1800 cm^{-1} to cover chromophores and carbohydrate-related bands. Photodegradation was controlled as described previously [4]. For the cotton samples a neutral density filter (ND 0.3) was used during 30 s of acquisition time. Background correction for the linear fluorescence was made and all spectra were normalised to the ~ 1375 cm^{-1} band. In the cases of which a quantitative analysis was performed, curve fitting using Lorentzian peak functions was applied.

RESULTS AND DISCUSSION

Model compounds experiments

UV-vis DR (*k/s*) and UV-RR (@ 325 nm) spectra of model compounds **1-7** deposited on cotton linters are depicted in Fig. 2. DR spectra revealed the maxima of absorption for all conjugated carbonyl structures represented by models **1-4** between 300 and 350 nm. An expected small bathochromic shift of around 5 nm was observed for syringyl units with respect to guaiacyl α-carbonyl models (comparing models **2** and **3**) due to electron donating effect of methoxyl substitute. The reaction products of *o*-quinone type (**5b**) arisen from oxidation of **5** with ClO_2 also exhibited a notable shoulder at ca. 325 nm in the DR spectrum though the contribution to the absorption at 325 nm of muconic acid type structures substituted with carbonyl-containing moieties can not be excluded as well (usually muconic acid structures absorb at ca. 260 nm). Hence the laser beam excitation at 325 nm used for acquiring the UV-RR spectra is an appropriate wavelength for selective detection of such kind of chromophore structures. The structures **1-5b** revealed the highest resonance enhancement in the UV-RR spectra at around 1600 cm^{-1} (Fig. 2). This finding is in tune with conclusions of previous work on the analysis of bleached eucalypt pulps by UV-RR (@ 325 nm) where the signal centred at ca. 1600

cm^{-1} was assigned to conjugated carbonyl structures both from residual lignin and carbohydrates [3]. Conjugated carbonyl structures belonging to oxidised residual lignin in pulp (quinones, etc. [5]) and to degraded carbohydrates contribute to pulp colour and brightness reversion.

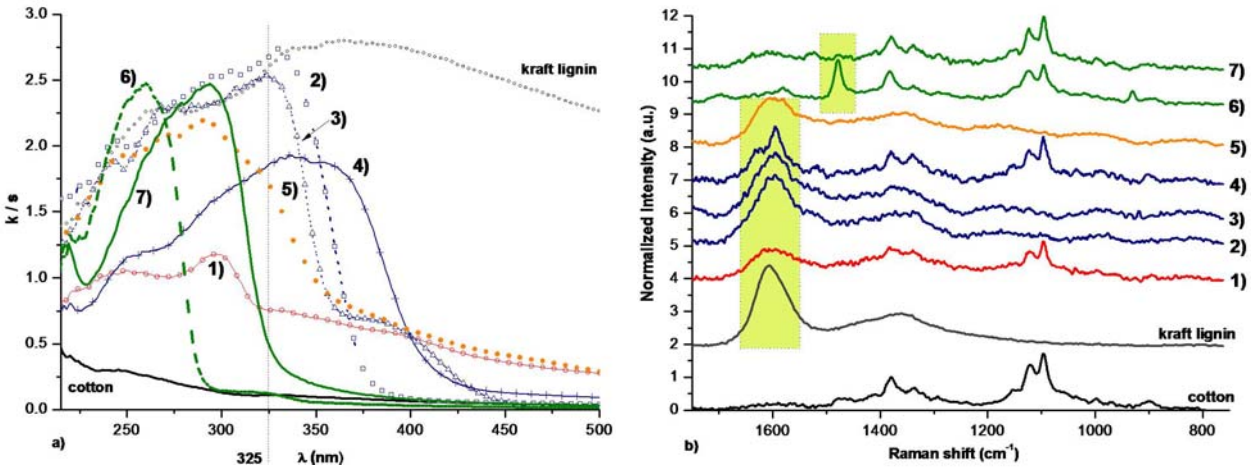


Figure 2. Spectra of the original cotton and after the deposition of kraft lignin and of the model compounds depicted in Fig.1: a) *k/s* spectra; b) UV-RR spectra @ 325 nm (offset spectra).

Kraft lignin deposited on cotton linters revealed the increased absorption across the UV-vis spectral region of 300 – 500 nm contributing significantly to absorbance at 457 nm where pulp ISO brightness is measured (ISO 3688/2470). The absorption maximum for kraft lignin was detected at around 365 nm and belongs to various conjugated structures, both containing conjugated carbonyl groups and polyunsaturated aromatic structures [5]. The intensity of Raman signal at ca. 1600 cm^{-1} from kraft lignin was even higher than for monomeric model compounds. Such high intensity has hindered a successful band deconvolution upon spectra fitting and led to eclipsing of carbohydrate-related bands at ca. 1120 and 1093 cm^{-1} assigned to stretching vibration modes of COC/OCO groups [3]. Similar features were observed for cotton samples with deposited models **2, 3** and **5a,b** possessing rather high absorption at 325 nm in *k/s* spectra (Fig. 2). Another reason for the disappearance of aforementioned cellulose bands at 1120 and 1093 cm^{-1} may be the specific micro-environment of cotton surface with deposited models. Nevertheless, these experiments confirm the potential of UV-RR at @ 325 nm laser beam excitation to assess minor amounts of chromophore moieties.

Models **6** and **7** belong to furan derivatives. Furan compounds are well-known carbohydrate degradation products and precursors of chromophores in pulp [7]. In kraft pulp bleaching, 2-furancarboxylic acid (**6**) and 5-formyl-2-furancarboxylic acid (**7**) are formed from HexA residues under acidic conditions [8]. Furthermore, furan derivatives were claimed as important contributors to pulp colour formation upon ageing [9]. The bathochromic shift of **7** with respect to **6** in UV-vis DR spectra is assigned to an extended conjugation effect of the former (Fig. 2). As a consequence the contribution to *k/s* at 325 nm of **7** is much superior to **6**. This explains the absence of signal at 1600 cm^{-1} for **6** in UV-RR spectrum and a weak signal for **7**. However, model **6** revealed the principal resonance signal at 1478 cm^{-1} though model **7** showed the peak at 1525 cm^{-1} in UV-RR spectrum (Fig. 2). Hence the signals at 1472-1478 cm^{-1} and at 1525 cm^{-1} in UV-RR spectra are characteristic for furan derivatives.

The characteristic Raman wavenumbers for the different chromophore moieties/pulp constituents are summarised in Table 1. The peak centred at ca. 1595 cm^{-1} in UV-RR@325 spectra is assigned basically to polyconjugated carbonyl structures though poly-unsaturated moieties contribute also to signals at ca. 1630 nm (model **4**). The signals at ca. 1470-1480 cm^{-1} are assigned to 2-furancarboxylic acid derivatives. Most of chromophores presented unresolved bands that could be separated by peak-fitting into several peaks. Hence the chromophore bands are overlapped and coupled of different vibration modes, therefore some shifting is expected in peaks centre, not to mention the effect of the

specific chemical micro-environment in pulps. The resolved band at 1599±2 cm^{-1} also belongs to the aromatic ring symmetric stretching [10]. The band at 1522 ± 2 cm^{-1} is always present in conjugated carbonyl structures **2-4** and tentatively assigned to their enolic configuration.

Table 1. Characteristic Raman wavenumbers of eventual chromophores in kraft pulps.

Chromophore system	Wavenumber of major signals (cm^{-1})	
	maximum intensity	peaks fitting[(a)]
Kraft lignin	1606	1602
1. *p*-quinone	1595	1616
2. 3,5-dimethoxy-4-hydroxyacetophenon	1594	1599; 1521
3. 4-hydroxy-3-methoxyacetophenon	1594	1599, 1523
4. 3,5-dimethoxy-4-hydroxy-cinnamic acid	1595	1630; 1594; 1522
5a muconic acid type and **5b** *o*-quinone derivatives + **3**	1596	1601; 1528
6. 2-furancarboxylic acid	1478	1700; 1579; 1478; 930
7. 5-formyl-2-furancarboxylic acid	1525	1606; 1525; 1472
Xylan	1606	1593
HexA-enriched xylan	1595	1592

(a) – deconvolution using Lorentzian peak functions

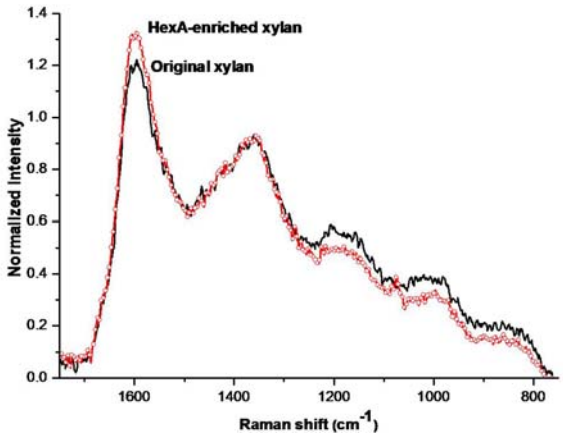


Figure 3. UV-RR spectra @ 325 nm of xylans.

The analysis of glucuronoxylan sample by UV-RR spectroscopy (Fig. 3; Table 1) reveals it as an important source of chromophores. The same feature was highlighted for xylans isolated from bleached eucalypt pulps [3]. Although xylan was isolated from peracetic holocellulose, some minor lignin-derived fragments (<1%) have survived, being chemically linked to xylan backbone. The maximum intensity at 1606 cm^{-1} for kraft lignin and xylan coincides (Table 1). Upon the harsh alkaline treatment of xylan aiming to convert GlcA residues to HexA moieties, the associated lignin also suffers structural changes thus increasing the intensity of chromophore band. Hence some increase of signal at 1606 cm^{-1} in HexA-enriched xylan is due to degraded lignin rather than to HexA residues. In effect, the specific band at 1657 cm^{-1} assigned to CH=C< stretching in HexA [1] was not detected (Fig. 3). This is explained by lack of resonance since the excitation at 325 nm used for the acquiring of UV-RR spectra is farther from HexA specific absorption at 235 nm, thus confirming the selectivity of the precise detection of highly conjugated structures.

Monitoring of pulp bleaching

The bleaching of *E. globulus* kraft pulps along DEDED and OQ(PO)DP sequences was monitored by UV-vis DR and UV-RR spectroscopy (Figs 4 and 5, respectively) and the changes in chromophore composition interpreted from the knowledge gained with model compounds and literature data [10, 11]. During bleaching the removal of chromophores across the entire UV-vis spectrum took place, which depended significantly on the type and the order of addition of bleaching chemicals (Fig. 4). Thus D_0 and O stages were effective in pulp delignification (decrease of *k/s* at 280 nm). However, O stage, as well as the PO stage, was inefficient to degrade structures absorbing below 270 nm, assigned mostly to HexA and lignin structures inaccessible to oxidation. Both E_1 (E_{OP}) and PO stages were decisive for the brightness gains (*k/s* values at 457 nm) due to elimination of oxidised lignin with conjugated carbonyl moieties (quinones, phenolic α–CO structures, etc.) arisen in previous D_0 and O

stages, respectively. The removal of chromophores after E_2D_2 (ca. 91% ISO brightness) stages of $D_0E_1D_1E_2D_2$ sequence, was insignificant in relation to D_1 (ca. 88 %) stage. In contrast, the D stage in OQ(PO)DP sequence was responsible for the massive release of chromophore structures of all origin. In particular, the significant elimination of HexA (decrease of *k/s* at 235 nm) and polyunsaturated structures (*k/s* at 300-400 nm) was observed (Fig. 4). The final P stage in OQ(PO)DP bleaching led to an ISO brightness gain of 4.5% and in contrast to final D stage of DEDED sequence, showed a notable elimination of conjugated carbonyl structures (decrease of *k/s* at 300-350 nm). The UV-RR spectra @325 nm revealed the levels of highly conjugated structures, assessed at ca. 1600 cm^{-1}, linearly correlated to *k/s* values at 325 nm of the UV-vis DR spectra (Fig. 6). It is important to note that the amount of chromophores detected at 325 nm not necessarily directly correlates to the ISO brightness values (measured at 457 nm) of pulps. The DEDED and OQ(PO)DP bleached pulps with the same ISO brightness level (ca 91%) possessed different amount of residual unsaturated structures at 325 nm: *k/s* values of 0.11 for DEDED and of 0.08 for OQ(PO)DP. A selective on-line monitoring of specific chromophores across the bleaching sequence using RR spectroscopy at different excitation wavelengths would be a useful tool for the quality control of pulp.

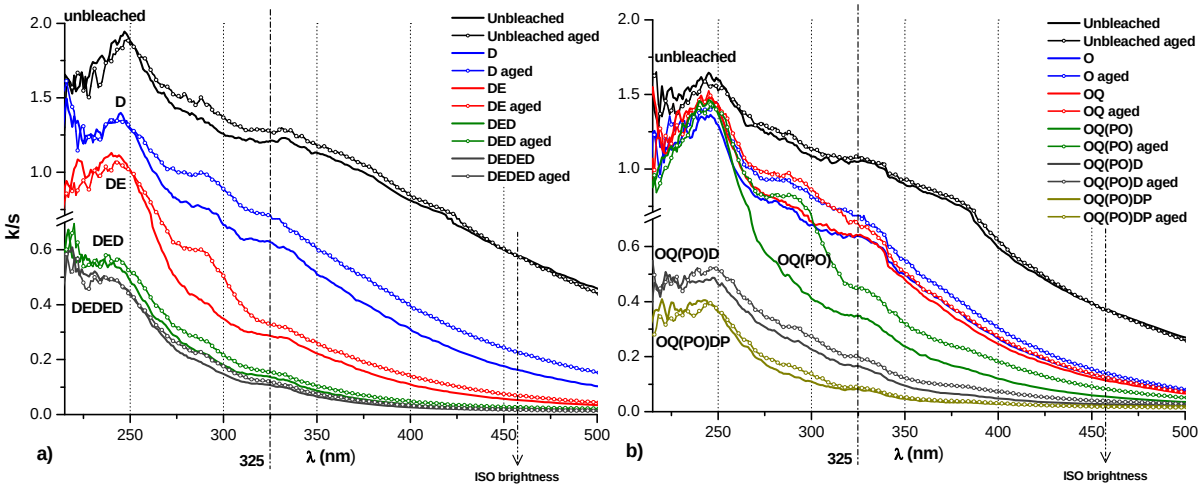


Figure 4. UV-vis DR spectra of *E. globulus* pulps throughout each ECF bleaching stage and following wet-thermal reversion: a) DEDED and b) OQ(PO)DP industrial sequences.

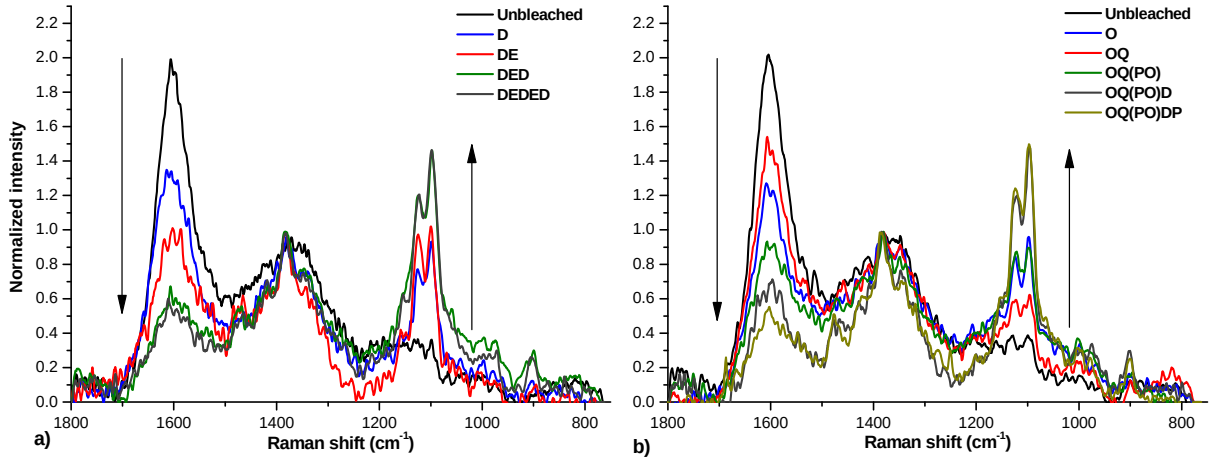


Figure 5. UV-RR spectra @ 325 nm of *E. globulus* pulps throughout ECF bleaching: a) DEDED and b) OQ(PO)DP industrial sequences.

The removal of chromophore structures during bleaching changes the specific chemical micro-

environment on fibre surface favouring better polysaccharides exposition. This explains, at least partially, the increase of signal at ca. 1120 and 1093 cm^{-1} while the signal from chromophore structures at ca. 1600 cm^{-1} decreases (Fig. 5). However, the signals from increased amounts of carbohydrate CO/COOH groups in pulps also contribute to the bands at ca. 1120 and 1093 cm^{-1} [3]. The excessive oxidation of polysaccharides in the final stages may be prejudicial for the brightness stability of the bleached pulp. In fact, all fully bleached pulps revealed remarkable signal at ca. 1480 cm^{-1} and 910 cm^{-1} in UV-RR spectra (Fig. 5) assigned, according to the model compounds experiments, to furoic acid derivatives – precursors of chromophores formation upon pulp ageing. These structures are derived from thermal hydrolysis of oxidised polysaccharides. It may be also proposed that the formation of new polysaccharide-derived chromophores is one of the reasons for the difficult brightness development in the final D stage.

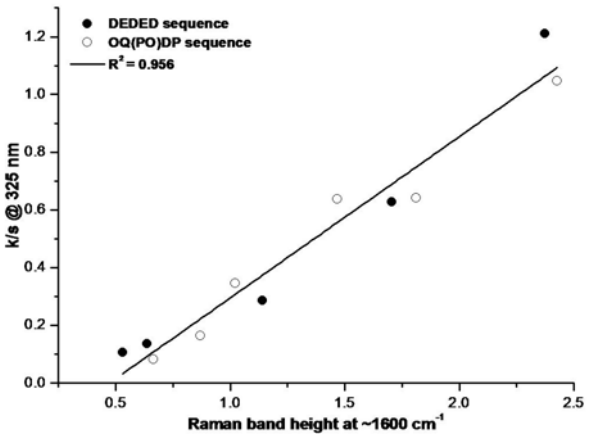


Figure 6. *k/s* @ 325nm *versus* 1600 cm^{-1} band height.

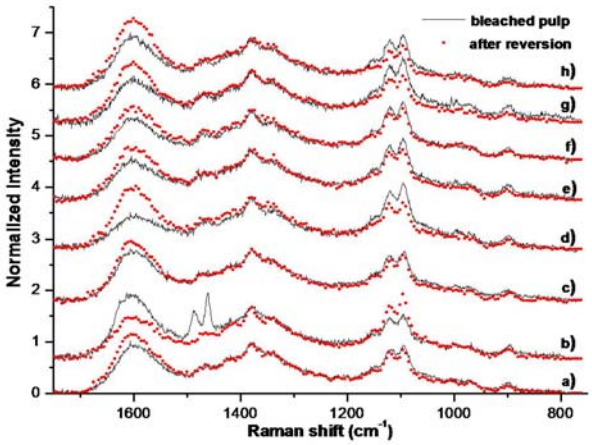


Figure 7. UV-RR spectra @ 325 nm of industrially produced *E. globulus* bleached pulps before and after reversion.

Brightness reversion

The appearance of new chromophore systems in pulps during wet-thermal ageing after different bleaching stages in DEDED and OQ(PO)DP sequences was assessed employing UV-vis DR spectroscopy (Fig. 4). Unbleached pulp and pulps after pre-bleaching stages (D_0/D_0E_1 and O/OQ(PO)) showed the decrease of absorption below 250 nm upon ageing with simultaneous rising of absorption above 250 nm through the visible region. Upon ageing, these pulps exhibited a notable broad band across 250-320 nm range with a maximum at ca. 290 as revealed by difference spectra (not shown). Besides residual lignin, a significant contribution to brightness reversion of these pulps may be expected from HexA that degrade to furoic acid derivatives giving rise to new chromophores such as reductic acid among others [9]. After D bleaching stages (D_2 in DEDED and D in OQ(PO)DP), new chromophores upon ageing were formed showing increased absorption above 230 nm (Fig. 4). The contribution of HexA to brightness reversion of these pulps is negligible (C_{HexA}< 2 mmol/kg odp) and, besides residual oxidised lignin, oxidised sugar residues absorbing at ≤ 230 nm may be hydrothermally degraded to quinone type derivatives (Theander products) [6]. In the final bleaching stages new unsaturated structures above 250 nm are formed because of thermal degradation of pulp components absorbing below 240 nm (Fig. 4). It was also shown previously that xylan isolated from fully DEDD bleached pulp revealed increased amount of conjugated structures [3]. The chromophore-referred band at ca. 1600 cm^{-1} in UV-RR spectra of DEDED and OQ(PO)DP bleached pulps was increased upon ageing. Hence the brightness reversion can be also monitored by UV-RR.

A series of industrial eucalypt kraft pulps fully bleached either by DEDD (pulps **a**, **c**, **e** and **h**) or by DEDED (pulps **b**, **d**, **f**, **g**) sequence, possessing different brightness reversion (0.65 and 0.29 PC number or 4.0 and 2.4 % ISO brightness loss, respectively) were analysed by UV-RR spectroscopy (Fig. 7). The increase of signal at ca. 1600 cm^{-1} is indicative of formation of new conjugated carbonyl structures upon ageing while simultaneous decrease of signals at ca. 1120 and 1093 cm^{-1} corroborates with thermal degradation of CO/COOH-containing structures from the polysaccharide component.

Alkaline extraction stage was suggested to be crucial to decrease the amount of unsaturated structures after D stage (see also Fig. 4) and to improve the brightness stability of the final bleached pulp. This also resembles the advantages of using a final alkaline P stage for the removal of reducing substances to prevent brightness reversion [12]. Therefore the omitted E_2 stage in DEDD bleaching led to worst brightness reversion of final pulp, when compared to DEDED pulp. Only pulp **b** in Fig. 7 (after D_2 in DEDED bleaching) showed a dissimilar behaviour upon reversion. This is probably related to the abnormally strong bands at 1486 and 1461 cm^{-1} assigned to furan derivatives, which were removed during ageing without their transformation into chromophores.

CONCLUSIONS

The UV-RR @325 nm spectroscopy was confirmed as a selective and powerful tool for the detection and quantification of chromophore structures in cellulosic pulps. The assignments of characteristic vibrations from particular chromophore structures have been performed using model compounds experiments thus allowing extending the UV-RR database. The signal at ca. 1600 cm^{-1} in UV-RR spectra of pulps linearly correlates to *k/s* at 325 nm of DR spectra fulfilling the conditions of Raman resonance for structures with conjugated carbonyl groups. The monitoring of eucalypt pulp bleaching in DEDED sequence by UV-RR allowed a proposition about formation of new polysaccharide-derived chromophores that hindered the brightness development in the final D stage. The last alkaline extraction stage in standard ECF DEDED bleaching was crucial to maintain high brightness stability of eucalypt kraft pulp due to removal of such chromophores.

Acknowledgements

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REFERENCES

1. M. Halttunen, J. Vyorykka, B. Hortling, T. Tamminen, D. Batchelder, A. Zimmermann, T. Vuorinen, Study of residual lignin in pulp by UV resonance Raman spectroscopy *Holzforschung*, **55**(6): 631-638 (2001). English
2. A. M. Saariaho, B. Hortling, A. S. Jaaskelainen, T. Tamminen, T. Vuorinen, Simultaneous quantification of residual lignin and hexenuronic acid from chemical pulps with UV resonance Raman spectroscopy and multivariate calibration *J. Pulp Pap. Sci.* , **29**(11): 363-370 (2003).
3. P. E. G. Loureiro, A. J. S. Fernandes, M. G. V. S. Carvalho, D. V. Evtuguin, The assessment of chromophores in bleached cellulosic pulps employing UV-Raman spectroscopy *Carbohyd. Res.*: (2010 doi: 10.1016/j.carres.2010.02.016).
4. D. V. Evtuguin, G. Rocha, B. J. Goodfellow, Detection of muconic acid type structures in oxidised lignins using 2D NMR spectroscopy *Holzforschung*, **63**(6): 675-680 (2009).
5. M. F. Pasco, I. D. Suckling, Chromophore changes during oxygen delignification of a radiata pine kraft pulp *Appita J.*, **51**(2): 138-146 (1998).
6. T. Rosenau, A. Potthast, P. Kosma, H. U. Suess, N. Nimmerfroh, Isolation and identification of residual chromophores from aged bleached pulp samples *Holzforschung*, **61**(6): 656-661 (2007).
7. O. Theander, D. A. Nelson, Aqueous, high-temperature transformation of carbohydrates relative to utilization of biomass *Adv. Carbohydr. Chem. Biochem.*, **46**: 273-326 (1988). English
8. A. Teleman, T. Hausalo, M. Tenkanen, T. Vuorinen, Identification of the acidic degradation products of hexenuronic acid and characterisation of hexenuronic acid-substituted xylooligosaccharides by NMR spectroscopy *Carbohyd. Res.*, **280**(2): 197-208 (1996).
9. O. Sevastyanova, J. Li, G. Gellerstedt, On the reaction mechanism of the thermal yellowing of bleached chemical pulps *Nord. Pulp Paper Res. J.*, **21**(2): 188-192 (2006).
10. U. P. Agarwal, "An overview of Raman spectroscopy as applied to lignocellulosics materials", *in*: D. S. Argyropoulos (ed.) Advances in Lignocellulosics Characterization, TAPPI PRESS, Atlanta, Georgia (1999).
11. A. Lahdetie, T. Liitia, T. Tamminen, A. S. Jaaskelainen, Reflectance UV-vis and UV resonance Raman spectroscopy in characterization of kraft pulps *Bioresources*, **4**(4): 1600-1619 (2009).
12. K. M. M. Eiras, J. L. Colodette, V. L. Silva, L. Barbosa, New insights on brightness stability of eucalyptus kraft pulp *Nord. Pulp Paper Res. J.*, **23**(1): 102-107 (2008).