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Short communication

Singlet oxygen-mediated degradation of lignin
Isolation of oxidation products from steam-exploded lignin from straw

Maurizio D'Auria*, Carlo Bonini, Lucia Emanuele, Rachele Ferri

Dipartimento di Chimica, Università della Basilicata, Via N. Sauro 85, 85100 Potenza, Italy

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Abstract

Four-hour irradiation of steam-exploded lignin from straw in the presence of singlet oxygen gave product showing high molecular weights. The only product obtained in low yield was the lactone **1**. Irradiation of the same lignin sample for longer times allowed to obtain higher quantity of the same compound. The largest amount of the lactone was obtained after 24 h irradiation. This behaviour can be explained considering that lignin from straw can be easily oxidised by singlet oxygen and that **1** can be obtained from the singlet oxygen oxidation of 5–5′ units in the lignin. © 2002 Elsevier Science B.V. All rights reserved.

Keywords: Singlet oxygen; Lignin; Photochemistry

1. Introduction

Lignin is a by-product in the paper industry and its treatment represents an important environmental problem. Two ways can be followed in order to obtain an environmentally sustainable development on this subject: the degradation and the use of this material as starting material for the synthesis of new ones. Considering lignin degradation, our attention has been devoted to the use of singlet oxygen.

In 1975, Gellerstedt and Petterson [1] proposed that singlet oxygen could be involved in the photochemical degradation of lignin. In particular, Gellerstedt and Petterson [1] proposed that singlet oxygen could have a role in the oxidation of some stilbene moieties in lignin. In 1984, Forrskåhl [2] made the hypothesis that singlet oxygen could be involved in photochemical degradation of lignin. In this case, singlet oxygen could be generated through a sensitised reaction from the carbonyl groups of the lignin itself. However, in 1980 Nimz and Turznik [3] found that singlet oxygen showed a poor reactivity with lignin model compounds. Singlet oxygen was generated through a well-known reaction of sodium hypochlorite with hydrogen peroxide. More recently, the same reaction for singlet oxygen production was used in order to obtain lignin degradation in pulp [4]. In this case, interesting results were obtained but they did not allowed the definition of a new degradation procedure.

Recently, we showed that lignin degradation can be obtained using photochemically generated singlet oxygen. In particular, we studied the behaviour of singlet oxygen towards lignin model compounds, showing that oxidation reactions occur on the phenoxy part of the molecule [5,6]. Furthermore, we found that the irradiation with visible light in the presence of both a sensitizer and oxygen can degrade isolated lignin from steam explosion and lignin in pulp from steam explosion [7–9].

The irradiation of a steam-exploded lignin from beech in the presence of oxygen gave a residue whose analysis showed the presence of vanilline and sinapyl alcohol [10]. On the contrary, the treatment for different irradiation times gave sinapyl alcohol, 4-hydroxy-3,5-dimethoxybenzaldehyde and 2,4-dioctylphenol [11].

In this paper, we want to report our results on the photochemical degradation of a sample of steam-exploded lignin from straw. Furthermore, in our study the degradation products were isolated, in order to determine the exact amount of the degradation products obtained, and identified by using spectroscopic methods.

2. Material and methods

The material used as source of lignin (straw) was mechanically reduced in pieces of about 1 cm of length and added with water to rise its initial content to the value of 50%. Steam explosion runs were carried in a 10 l batch

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* Corresponding author. Tel.: +39-971-202240; fax: +39-971-202223.
E-mail address: dauria@unibas.it (M. D'Auria).

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reactor, loading about 0.5 kg of material each cycle. Treatment conditions were 188 °C and 3 min with $\log R_0 = 3.2$. The raw material (100 g) was extracted two times with hot water (250–300 ml, 65 ± 5 °C) in order to eliminate sugars and hemicelluloses. The extraction of lignin from exploded materials has been carried out by 1.5% sodium hydroxide solution (250 ml) at 90 °C in 15 min for two times. Lignin was precipitated at pH 2 with 20% H_2SO_4 when the solution was still warm, filtered, washed and dried at 105 °C.

Elemental analyses were obtained with a Carlo Erba Elemental Analyzer 1106. ^{13}C NMR spectra were recorded with a Bruker 300 AM instrument. All the spectra were recorded in DMSO-d_6. Gel permeation chromatography analyses were performed on a Hewlett-Packard HPLC by using H-P Plgel 5 μm column. The lignin samples were acetylated (acetic anhydride and pyridine) before the use. THF was used as mobile phase. Spectrophotometric-grade THF was used and distilled (oven $LiAlH_4$) before the use. The chromatograms were obtained using an UV detector at 280 nm. The conversion from elution time to molecular weight was performed by using a calibration obtained by using polystyrene samples [12]. Cary 2300 spectrophotometer was used for the UV spectra. Spectrophotometric-grade 1:1 acetonitrile–methanol mixture was used as solvent.

2.1. Characterisation of lignins from steam explosion

The results of elemental analyses were: C, 59.19%, H, 6.12%, N, 1.39% and S, 0.46%. We analysed the presence of carbon and hydrogen in order to characterise the lignin, but also the presence of both nitrogen as a marker of the presence of proteic materials in the lignin and sulphur as a marker of the presence of sulphonated lignins. The elemental analysis allows us to give a molecular formula expressed in phenylpropanoid (C-9) units of $C_9H_{11.17}O_{7.66}$ corresponding to the molecular weight of 242.

The distribution of acetylated lignins considering their molecular weights was obtained by using gel permeation chromatography. The results are depicted in Fig. 1. Our lignin sample has $\bar{M}_n = 807$, $\bar{M}_w = 3486$, $\bar{M}_n = 807$ and $\bar{M}_z = 14\,292$. The UV spectra of our samples were recorded in 1:1 acetonitrile–methanol. We recorded also the differential spectra obtained carrying out the spectrum of the samples in 1 M NaOH vs the standard solution. These data allow us to give the amount in meq g^{-1} of some structural features in the lignin samples. In this case, we were able to give the amounts of syringyl and guaiacyl phenols (Type I) (15.74 meq g^{-1}) [13]. In this case, our lignin showed absorptions at $\lambda = 204$ nm ($D = 267.72\,l\,g^{-1}\,cm^{-1}$), $\lambda = 222$ nm (shoulder, $D = 118.11\,l\,g^{-1}\,cm^{-1}$), $\lambda = 280$ nm ($D = 51.18\,l\,g^{-1}\,cm^{-1}$), $\lambda = 302$ nm (shoulder, $D = 31.50\,l\,g^{-1}\,cm^{-1}$) and $\lambda = 330$ nm ($D = 17.24\,l\,g^{-1}\,cm^{-1}$). The FTIR spectrum of lignin showed absorptions at 2920, 2852, 1702, 1655, 1518, 1511, 1463, 1422, 1253, 1223 and 1121 cm^{-1}.

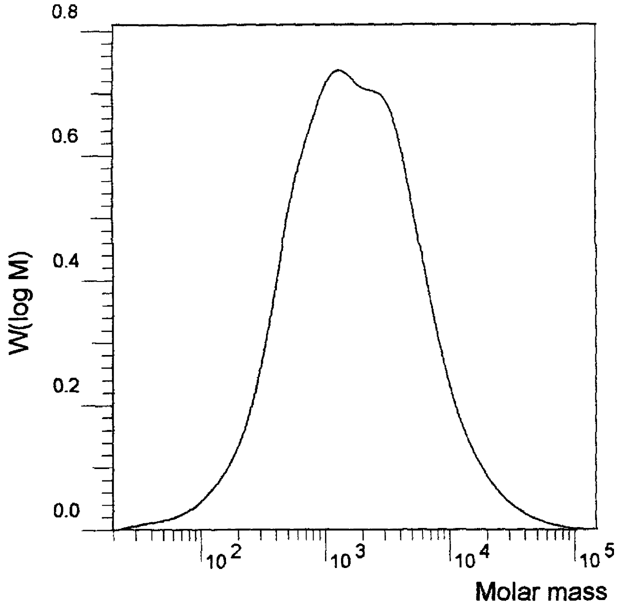

Fig. 1. Molecular weight distribution of steam-exploded lignin from straw ($R_0 = 3.2$).

Finally, the characterisation of the lignins used in this work was completed by using the ^{13}C NMR spectroscopy. With our sample, we observed signals at δ (DMSO-d_6) 152, 147.5, 138, 134.5, 115, 104, 86, 72.5, 60, 55.9 and 40 ppm [14,15]. From these data, we can observe the presence of guaiacyl and syringyl structures. These structures are both β-O-4 etherified and non-etherified. We observe the presence of both *threo* and *erithro* structures. We note the presence of structures of type cinnamaldehyde and stilbenes. The ^1H NMR spectrum of acetylated sample of lignin in $CDCl_3$ showed signals at δ 1.22, 1.7, 2.0, 2.1, 2.2, 2.3, 2.6, 3.7, 6 and 6.5–7.3 ppm.

2.2. Reactions with singlet oxygen

A solution of the lignin (100 mg) in a 1:1 mixture of acetonitrile–methanol (10 ml) containing 5×10^{-4} M Rose Bengal was irradiated in a Pyrex tube surrounded by a Pyrex water-jacket connected to a Haake D9-G thermostat to maintain the temperature at 13.0 ± 0.1 °C. The Pyrex tube was dipped into a 1% (w/v) solution of $NaNO_2$ in order to cut-off the irradiation at 400 nm. The solution was previously saturated with bubbling oxygen for 1 h. The irradiation was performed by using a 50 W tungsten–halogen lamp. The solvent was evaporated and the residue was chromatographed on silica gel eluting with 3:7 MeOH–$CHCl_3$. The collected fractions were analysed.

Mass spectra were obtained with a Hewlett-Packard 5971 mass-selective detector on a Hewlett-Packard 5890 gas chromatograph (OV-1 capillary column between 70 and 250 °C (20 °C min^{-1})).

3. Results and discussion

To perform our experiments, we used a steam-exploded lignin from straw. The characterisation of this lignin showed that in the structure of this lignin only guaiacyl and syringyl units are present. This result was obtained from the analysis of ^{13}C NMR spectrum. Elemental analysis showed the presence of a little amount of proteic material while the presence of sulphur is not relevant. However, elemental analysis shows that this sample is highly oxidised with a large amount of oxygen in the molecular formula. In fact, the presence of oxygen is larger than in other samples of steam-exploded lignins we used in the past [7,10]. In agreement with this statement, the ^{1}H NMR spectrum showed the presence of signals due to the presence of aromatic acetates, and the differential UV spectrum confirms the presence of large amount of phenolic hydroxy groups in the structure.

Gel permeation chromatography showed that most of the molecules of our sample has a molecular distribution in the range 100–100 000 with $\bar{M}_w = 3486$. This datum confirms the evidence that the steam explosion process induces a strong destructuration in the lignin structure giving samples with relatively low molecular weight.

We carried out the irradiation of a lignin sample in 1:1 acetonitrile–methanol for 4 h in the presence of oxygen and Bengal Rose. The residue after evaporation of the solvent was chromatographed on silica gel. We obtained several chromatographic fractions whose GC–MS analyses showed that lignin fragments are present. Only one fraction gave a product in low yields (6 mg, Table 1). This compound showed an MS spectrum with peaks at *m/z* 230 (relative abundance: 8%), 229 (51), 228 (79), 212 (9), 211 (53), 185 (25), 171 (10), 143 (10), 129 (32), 115 (11), 111 (11), 103 (13), 102 (84), 98 (13), 97 (20), 87 (15), 85 (24), 84 (12), 83 (21), 73 (43), 71 (33), 69 (30), 61 (26), 60 (76), 59 (24), 57 (51), 56 (14), 55 (47), 44 (10), 43 (100), 42 (18) and 41 (53). Then, assuming a molecular weight of 229, the most significant fragmentation in the spectrum were M-18 (*m/z* 211), M-44 (*m/z* 185), M-100 (*m/z* 129) and M-127 (*m/z* 102). The infrared spectrum showed absorptions at $\nu_{\max}$ 2960, 2930, 2860, 1745, 1610, 1520, 1470, 1390, and 1115 cm^{-1}. The spectrum showed absorption at 1745 cm^{-1} that can be attributed to an ester or lactone. The ^{1}H NMR spectrum (DMSO-d$_6$) showed a singlet at δ 4.35 ppm (1H), a signal at δ 3.57 ppm (singlet, 8H) and a large singlet at δ 3.18 ppm (2H). The ^{13}C NMR spectrum of the isolated compound showed signals at δ 175.3, 130.3, 129.9, 128.4, 70.4, 61.7, 61.4, 59.9, 39.8 and 29.6 ppm. These data are in agreement with the formation of compound **1** (Fig. 2). In fact, this structure is in agreement with the presence of the absorption at 1745 cm^{-1} in the infrared spectrum. Furthermore, the signal at δ 175.3 ppm in the ^{13}C NMR spectrum can be attributed to the carbonyl carbon in the lactone, while the peaks at δ 130.3 and 129.9 ppm can be assigned to the olefinic carbons. The signals at δ 70.4, 61.7 and 61.4 ppm in the same spectrum can be assigned to the C-2, C-1 and C-5, respectively. The signal at δ 59.9 ppm can be identified with the methoxy groups. Finally, the signals at δ 39.8 and 29.6 ppm can be assigned to C-7 and C-6, respectively. The ^{1}H NMR spectrum can be explained assuming that the singlet at δ 4.35 ppm is due to the proton at C-2, that the signal at δ 3.37 ppm to both CH$_3$O and C-7 protons, while the signal at δ 3.18 ppm can be attributed to C-6 protons. Compound **1** has molecular weight 458: considering that **1** is a dimeric structure we wait for a molecular peak at *m/z* 229 in agreement with the observed mass spectrum. Furthermore, the peak at M-18 can be attributed to a loss of water from an alcoholic function. The signal at *m/z* 185 can be due to the loss of CO$_2$, while that at *m/z* 129 can be attributed to a fragment after the loss of the fragment **2** (Fig. 3). Finally, the peak at *m/z* 102 can be attributed to the ion **3** (Fig. 3).

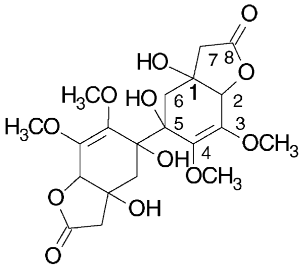

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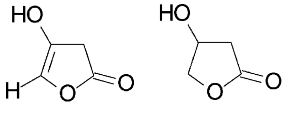
Fig. 2. Structure of the compound isolated in the residue after singlet oxygen-mediated degradation of lignin from straw.

Table 1
Singlet oxygen-mediated degradation of lignin from straw: isolation of residual product

Lignin (mg)	Irradiation time (h)	Product (mg)
103	4	6
103	8	8
102	12	24
102	24	28
102	48	24
101	144	3

On the basis of this result, we carried out lignin degradation for different irradiation time (Table 1). The irradiation of the same sample of lignin for 8 h gave almost the same amount of **1**, while lignin polymers form the most abundant fraction of the residue. On the contrary, when the irradiation was carried out for 12, 24 and 48 h, we observed that **1**



m/z 100 **2**   *m/z*102 **3**

Fig. 3. Some ions and fragments observed in the mass spectrum of **1**.

can be obtained in larger amounts (Table 1). The best result we had was obtained after 24 h irradiation with 28% yield (w/w) of compound **1**. Prolonged irradiation time caused lower yield of **1** (Table 1): in fact, after 6 days of irradiation, only 3 mg of **1** was isolated in the reaction mixture.

It is interesting to note that, in this case, we was not able to isolate lignin monomers such as sinapyl alcohol and 4-hydroxy-3,5-dimethoxybenzaldehyde as reported in our previous works [10,11]. Probably, the different behaviour observed depends on several factors. First, in our previous works we used steam-exploded lignin from beech, while in this work we used steam-exploded lignin from straw. Lignin from straw usually shows larger amount of phenolic hydroxy groups: in fact, while our sample showed 15.74 $\mathrm{meq\,g^{-1}}$ of syringyl and guaiacyl phenols (Type I), lignin from beech we used in our previous works showed 1.54 $\mathrm{meq\,g^{-1}}$ of Type I phenols, 0.13 $\mathrm{meq\,g^{-1}}$ of Type II phenols (hydroxycinnamic alcohol) and 0.11 $\mathrm{meq\,g^{-1}}$ of Type IV phenols (stilbenic phenols). Then, lignin from straw showed 10 times more phenols than that from beech. Phenols can be more easily oxidised by singlet oxygen. The oxidation can induce extensive destruction of the aromatic part of the polymer.

We observed a product deriving from less oxidable 5–5′ dimeric structures. The 5–5′ junction is not destroyed and singlet oxygen can only attach the ring to give the alcoholic functions at the 5 and 5′ positions. Singlet oxygen can also attack the aromatic ring to give an 1,2-endoperoxide and can oxidise a terminal alcoholic function to acid. This acid attacks the endoperoxide to give the corresponding lactone. In conclusion, we found that the irradiation of steam-exploded lignin from straw in the presence of both oxygen and singlet oxygen sensitizer allowed us to obtain acceptable amount of a compound never described before. This result confirms the hypothesis that oxidative degradation of lignin can be considered a useful method to obtain interesting fine chemicals.

References

[1] G. Gellerstedt, E.-L. Petterson, Acta Chem. Scand. B 29 (1975) 1005.
[2] I. Forrskåhl, J. Photochem. 25 (1984) 197.
[3] H.H. Nimz, G. Turznik, Cellulose Chem. Technol. 14 (1980) 727.
[4] J. Szabo, M. Fiserova, M. Kosik, Cellulose Chem. Technol. 28 (1994) 183.
[5] C. Crestini, M. D'Auria, J. Photochem. Photobiol. A 101 (1996) 69.
[6] C. Crestini, M. D'Auria, Tetrahedron 53 (1997) 7877.
[7] C. Bonini, M. D'Auria, L. D'Alessio, G. Mauriello, D. Tofani, D. Viggiano, F. Zimbardi, J. Photochem. Photobiol. A 113 (1998) 119.
[8] C. Bonini, M. D'Auria, G. Mauriello, D. Viggiano, F. Zimbardi, J. Photochem. Photobiol. A 118 (1998) 107.
[9] G. Bentivenga, C. Bonini, M. D'Auria, A. De Bona, G. Mauriello, Chemosphere 39 (1999) 2409.
[10] G. Bentivenga, C. Bonini, M. D'Auria, A. De Bona, J. Photochem. Photobiol. A 128 (1999) 139.
[11] G. Bentivenga, C. Bonini, M. D'Auria, A. De Bona, G. Mauriello, J. Photochem. Photobiol. A 135 (2000) 203.
[12] G. Gellerstedt, Gel permeation chromatography, in: S.Y. Lin, C.W. Dence (Eds.), Methods in Lignin Chemistry, Springer, Berlin, 1992, pp. 487–497.
[13] S.Y. Lin, Ultraviolet spectroscopy, in: S.Y. Lin, C.W. Dence (Eds.), Methods in Lignin Chemistry, Springer, Berlin, 1992, pp. 217–232.
[14] R. Mörck, K.P. Kringstad, Holzforshung 39 (1985) 109.
[15] H. Nimz, D. Robert, O. Faix, L. Ramsey, Holzforshung 35 (1981) 16.