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GC-MS and IR spectroscopic analyses of the lignin-derived products from softwood and hardwood treated in supercritical water

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Abstract Softwood (*Cryptomeria japonica*) and hardwood (*Fagus crenata*) were treated in supercritical water (380°C, 100MPa) for 8s. The treated woods were fractionated to the water-soluble portion, methanol-soluble portion, and methanol-insoluble residues. For the methanol-soluble portion, which mainly consisted of lignin-derived products, gel permeation chromatography (GPC) and gas chromatographic-mass spectrometric (GC-MS) analyses were conducted to clarify the molecular weight distribution and to identify the monomeric products, respectively. GPC analysis revealed that the methanol-soluble portion contains monomeric and some oligomeric products. GC-MS analysis identified 19 guaiacyl compounds in the methanol-soluble portion from softwood, and 15 syringyl monomeric compounds in the methanol-soluble portion from hardwood. The structures of identified products included not only phenyl propane (C_6-C_3) units but also C_6-C_2 and C_6-C_1 units. In addition, the infrared spectra suggested that the methanol-soluble portion maintains the typical structure of lignin, although it is rich in condensed-type linkages with some changes in the propyl side chain. These results indicate that the supercritical water treatment cleaves not only ether linkages but also part of the propyl chains in lignin to give various aromatic compounds.

Key words Chemical conversion · FT-IR · GC-MS · Lignin · Supercritical water

Introduction

At present, large amounts of our energy, organic chemicals, and synthetic organic polymeric materials are obtained from fossil resources such as petroleum and natural gas.

However, fossil resources will be exhausted eventually, and carbon dioxide levels in the environment will increase through their combustion. On the other hand, lignocellulosic biomass is abundant and does not contribute to increases in carbon dioxide levels in the environment because it is carbon neutral. Therefore, utilization of biomass as a substitute for fossil resources must receive further attention.

Bobleter¹ proposed hydrothermal treatment of lignocellulosics without using any catalyst, and reported excellent conversion. In our research, not only subcritical (hydrothermal) but also supercritical fluid technologies have been applied to lignocellulosics to obtain bioenergy and useful chemicals.^{2–11} As a result, we have hitherto reported that lignocellulosics can be separated to carbohydrate-derived and lignin-derived products by supercritical water treatment.³ The former products, which mainly consist of polysaccharides, oligosaccharides, monosaccharides, and their decomposed products, have potential as a feedstock for ethanol fermentation.^{2,9,11,12} The lignin-derived products, which mainly consist of monomeric and oligomeric materials, may be useful substitutes for the chemicals from fossil resources.^{8,10,11} To explore these possibilities, characterization of lignin-derived products from supercritically treated lignocellulosics is of importance.

On the other hand, some research has shown that some isolated lignins, such as organosolv lignin and sulfonated lignin, can be treated by supercritical water to obtain useful chemicals and information on the degradation mechanism of lignin.^{13–16} However, any isolated lignin has already been, in itself, modified during the isolation process. In addition, contamination with chemical agents in isolated lignins is inevitable. These facts often make it difficult to utilize lignin-derived products and to elucidate the degradation mechanism of native lignin. Therefore, characterization of lignin-derived products from wood itself is also important.

In our previous report, lignin-derived products from wood treated in supercritical water were analyzed. As a result, it was clarified that the lignin-derived products mainly consist of condensed-type linkages of lignin due to

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the preferential degradation of the ether linkages during supercritical water treatment.^{8,11} In addition, monomeric and dimeric lignin-derived products from softwood lignin were identified with analysis by gas chromatography-mass spectrometry (GC-MS).^{10,11}

However, GC-MS analysis of lignin-derived products from hardwood has not been conducted. Lignins from softwood and hardwood differ somewhat in composition with methoxyl substitution. The former is dominated by guaiacyl nuclei, but the latter is composed by guaiacyl and syringyl units. Thus, we must consider these diversities of lignin during an investigation. Furthermore, the overall characterization of the lignin-derived products cannot be carried out by GC-MS alone.

In this study, therefore, lignin-derived products from softwood and hardwood treated in supercritical water were subsequently characterized by gel permeation chromatography (GPC) and GC-MS analyses. In addition, infrared spectroscopic analysis was also conducted to obtain information on its potential for efficient utilization and its degradation behavior.

Materials and methods

Samples and chemicals

Japanese cedar (*Cryptomeria japonica*) and Japanese beech (*Fagus crenata*) flours (80 mesh pass; <315 μm) were used as soft-wood and hardwood samples, respectively. Distilled water used for supercritical water treatment and methanol used for fractionation were of high performance liquid chromatography (HPLC) grade, while other chemicals were reagent grade and were used without purification. 2,2'-Dihydroxy-3,3'-dimethoxy-5,5'-dimethyl biphenyl was prepared according to the method by Kratzl and Vierhapper.¹⁷ The milled wood lignins (MWL) from Japanese cedar and beech were prepared according to the method by Bjorkman.¹⁸

Supercritical water treatment and fractionation of the methanol-soluble portion

Supercritical water treatment and fractionation were conducted in the same manner as described in our previous reports.^{8,10} Water (4.9 ml) was fed with 150 mg of wood flour to a batch-type reaction vessel made of Inconel-625.¹⁹ It was quickly heated by immersing the reaction vessel in a molten tin bath preheated at an appropriate temperature. To stop the reaction, the reaction vessel was immersed in a water bath. During this treatment, the temperature and pressure in the reaction vessel were monitored by a thermocouple and a pressure gauge attached to the reaction vessel. In this study, the sample was treated under the supercritical water condition of 380°C and 100 MPa for 8 s.

After supercritical water treatment, 4.9 ml of water-soluble portion was separated from the water-insoluble portion by filtration. The water-insoluble portion was then

treated with 10 ml of methanol for 10 min by sonication and separated into the methanol-soluble portion and the methanol-insoluble residue by centrifugation. This methanol-soluble portion was used for analytical study. The methanol-insoluble residue was dried at 105°C and weighed to obtain the yield.

Analytical methods

The lignin content in wood was determined as the sum of Klason lignin and acid-soluble lignin.²⁰

The molecular weight (MW) distribution of the methanol-soluble portion and the methanol-insoluble residue was estimated by GPC on a HPLC system (Shimadzu LC-10A) with a refractive index detector. Shodex KF-801, KF-802, KF-802.5, and KF-803 columns (Showa Denko, Tokyo, Japan) were connected in tandem. Tetrahydrofuran was used as the carrier solvent at 0.6 ml/min. The methanol-insoluble residues from Japanese cedar and Japanese beech could not be completely dissolved in any solvent, but 20% and 23% could be dissolved in dimethylsulfoxide (DMSO), respectively. Thus, the methanol-soluble portion and DMSO-soluble part of the methanol-insoluble residue were directly injected to the HPLC system.

Hitachi G-7000M and M-9000 gas chromatograph three-dimensional quadrupole mass spectrometer was used to identify the monomeric products in the methanol-soluble portion. CP-Sil 8 CB-Low Bleed/MS (30 m long, 0.25 mm inner diameter, 0.25 μm film thickness) was used as a capillary column. The temperature program was 1.0 min at 40°C, 5.0°C/min to 300°C and 1.0 min at 300°C. Helium was used as the carrier gas at a flow rate of 1.5 ml/min. Injector and detector temperatures were both 230°C. One microliter of the methanol-soluble portion was directly injected. Lignin-derived monomeric products were identified from their mass spectra by comparing their retention times with those of authentic compounds or with a help of the literature.^{21,22}

Fourier transform-infrared (FT-IR) spectra were recorded using a Shimadzu IR-8000 spectrophotometer using the KBr pellet technique.

Results and discussion

Table 1 shows lignin content and yield of the fractionated portions of Japanese cedar and beech as treated in supercritical water. It is already known that lignin is mainly collected in the methanol-soluble portion and methanol-insoluble residue.³ The yields of methanol-soluble portion from Japanese cedar and beech were 25.6% and 19.8%, respectively. Considering that lignin content of Japanese cedar and beech were 30.1% and 25.6%, respectively, about 80% of lignin in wood was isolated in the methanol-soluble portion after the supercritical water treatment. The residual lignin was mainly isolated in the methanol-insoluble residue,³ suggesting that contamination of

Table 1. Lignin content and yields of fractionated portions of Japanese cedar and beech treated in supercritical water (380°C, 100MPa) for 8 s

Sample	Lignin content (%)	Yields (%)		
		Water-soluble	Water-insoluble	
			Methanol-soluble	Methanol-insoluble
Japanese cedar	30.1	66.4	25.6	8.0
Japanese beech	25.6	72.7	19.8	7.5

lignin-derived products in the water-soluble portion was not very high.

Bobleter and Concin²³ reported degradation of hardwood and softwood lignin by subcritical water treatment (hydrothermal treatment) at a temperature of 372°C and density of 0.56 g/cm³.²³ They claimed that softwood lignin is less susceptible to decomposition by hydrothermal treatment than hardwood lignin. In our previous study, the same trend was observed.⁸ However, in the case of supercritical water treatment, there was no significant difference in lignin degradation rates for softwood and hardwood. This result was observed in our previous study⁸ and in the present data (Table 1). This is an interesting finding, and suggests that softwood lignin can be degraded in a similar fashion to hardwood lignin by supercritical water treatment. More detailed study, such as a morphological observation and model experiment, will be needed to explain this finding.

Figure 1 depicts GPC chromatograms of the methanol-soluble portions and the methanol-insoluble residues from Japanese cedar and beech as treated in supercritical water. Both methanol-soluble portions eluted from 45 min to 70 min. On the other hand, the elutions of methanol-insoluble residues were earlier than those of the methanol-soluble portions. The polystyrene standards with molecular weights (MW) of 1940, 1270, 925, 580, 162, and 102 were eluted at 48.5, 51.7, 54.0, 57.2, 66.0, and 70.5 min, respectively. In addition, lignin model compounds, such as 2,2', 3,3'-tetramethoxy-5,5'-dimethyl biphenyl, coniferylalcohol, acetoguaiacone, vanillin, and guaiacol, were eluted at 58.3, 60.6, 63.0, 63.9, and 65.6 min, respectively. The elution times of vanillin (MW 152) and guaiacol (MW 124) were shorter than that of the polystyrene standard (MW 162), indicating that the lignin model compounds tended to be eluted earlier than the polystyrene standards. Thus, the molecular weight distribution of the methanol-soluble portion was not underestimated by calibration with the polystyrene standards.

Based on the results of GPC analysis, Table 2 shows the molecular weight distribution of the methanol-soluble portions. The number-average MW, weight-average MW, and the polydispersity of the methanol-soluble portion from Japanese cedar were 335, 663, and 1.98, respectively, and were found to be smaller than those of the methanol-insoluble residue. The corresponding values for the methanol-soluble portion from Japanese beech were slightly smaller than those from Japanese cedar. Furthermore, the values are close to or lower than those for hydrothermally

Table 2. Molecular weight distribution of the methanol-soluble portion and the methanol-insoluble residue from Japanese cedar and beech treated in supercritical water

Sample	M_n	M_w	M_w/M_n
Methanol-soluble			
Japanese cedar	335	663	1.98
Japanese beech	333	569	1.80
Methanol-insoluble			
Japanese cedar	563	1178	2.09
Japanese beech	547	1162	2.12

M_n , number-average molecular weight; M_w , weight-average molecular weight; M_w/M_n , polydispersity

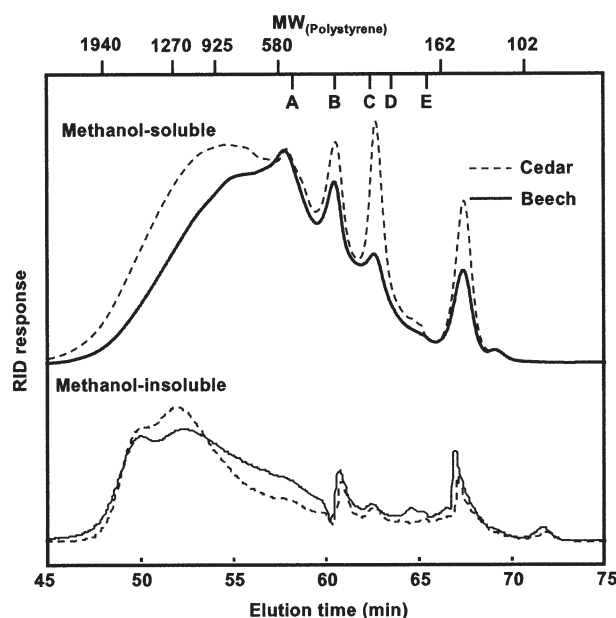


Fig. 1. Gel permeation chromatograms of the methanol-soluble portions and methanol-insoluble residues from Japanese cedar and Japanese beech treated in supercritical water. Labels A, B, C, D, and E represent the elution times of 2,2',3,3'-tetramethoxy-5,5'-dimethylbiphenyl (MW 302), coniferylalcohol (MW 180), acetoguaiacone (MW 166), vanillin (MW 152), and guaiacol (MW 124), respectively. The elution profile of the methanol-insoluble residue corresponded to the DMSO-soluble portion of the methanol-insoluble residue. MW, molecular weight; RID, refractive index detector

degraded lignin.²⁴ These results indicated that the methanol-soluble portions consist of lignin-derived monomeric, dimeric, trimeric, and some oligomeric products, but the methanol-insoluble residue might consist of larger MW

products than the methanol-soluble portion. In addition, the area after 60 min in Fig. 1, which is after the elution of coniferylalcohol, comprised about 35% of the total area, suggesting that the methanol-soluble portions should contain about 35% of monomeric lignin-derived compound.

Figure 2 shows the total ion chromatograms of the methanol-soluble portions from Japanese cedar and beech treated in supercritical water. The numbers in Fig. 2 represent the products as listed in Table 3. The results for Japanese cedar were reported as part of our previous work,¹⁰ and are tabulated here for comparison. The identified products from Japanese cedar had guaiacyl nuclei, and those from Japanese beech had both guaiacyl nuclei and syringyl nuclei.

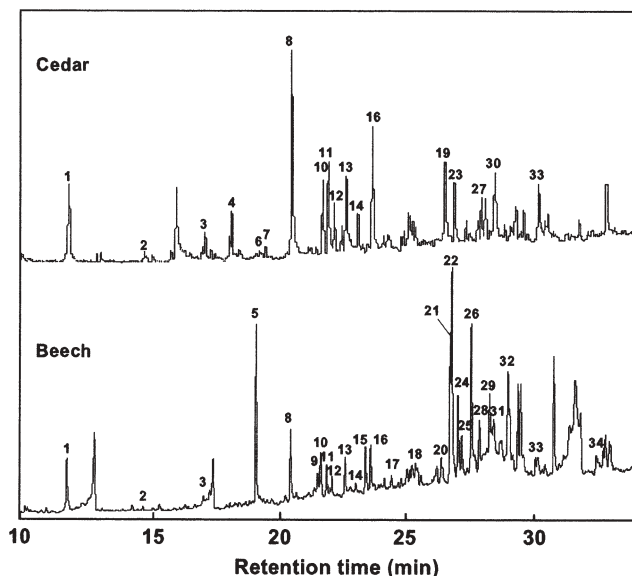
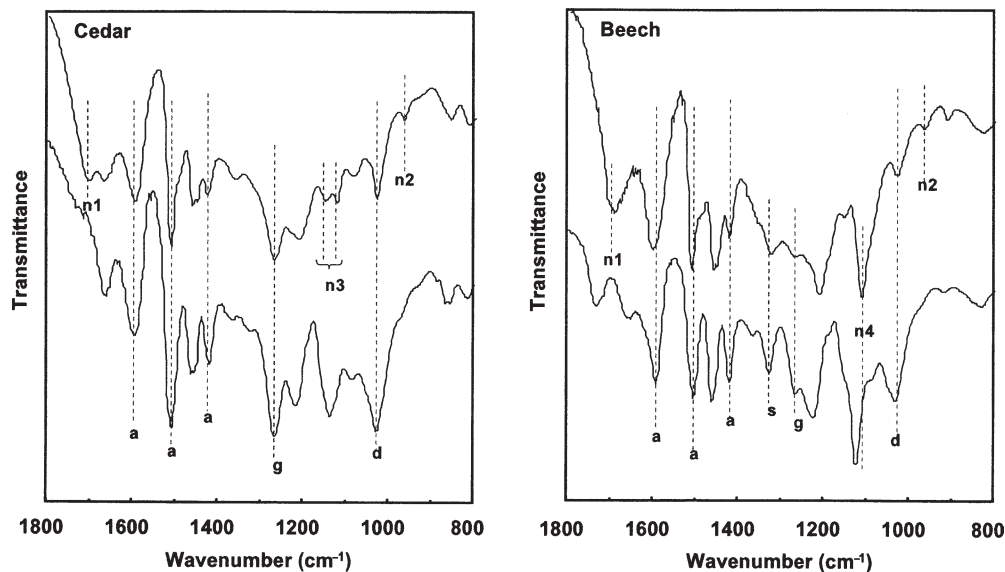


Fig. 2. Total ion chromatograms of the methanol-soluble portions from Japanese cedar and Japanese beech treated in supercritical water. The numbers on the peaks refer to the products listed in Table 3

Fig. 3. Fouries transform-infraced spectra of the methanol-soluble portions and milled wood lignins (MWL) from Japanese cedar and beech treated in supercritical water. Upper and lower spectra correspond to the methanol-soluble portion and MWL, respectively. For peak symbols, refer to text



Products **7, 8, 10, 12, 14, 16, 18, 19, 20, 22, 24, 25, 26, 27, 28, 30, 31, 32, 33,** and **34** (see Table 3) were composed of phenylpropane units (C_6-C_3). In our previous studies,^{8,11} it was demonstrated that the β -*O*-4 linkage of lignin was easily cleaved in supercritical water treatment. Thus, it is reasonable to assume that these products are derived from the cleavage of ether linkages of lignin.

In addition to C_6-C_3 units, C_6-C_2 and C_6-C_1 units were also detected. The former were ethylguaiaicol (**3**), vinylguaiaicol (**4**), homovanillin (**11**), acetoguaiaicone (**13**), ethylsyringol (**15**), vinylsyringol (**17**), homovanillic acid (**23**), and acetosyringone (**29**). The latter type were methylguaiaicol (**2**), vanillin (**6**), methylsyringol (**9**), and syringaaldehyde (**21**). The same products were also detected by GC-MS in hydrothermolysis (subcritical water condition) products from wood.²⁵⁻²⁷ In our previous studies of lignin model compounds treated in subcritical water and supercritical water,^{8,11} bond cleavage between C_β and C_γ (C_β/C_γ) in the C_6-C_3 unit was observed. Dealkylation of 2-isopropylphenol in supercritical water has also been reported.²⁸ Therefore, the existence of these products suggests that the cleavage of C_β/C_γ and C_α/C_β linkages of lignin take place in supercritical water and in subcritical water.

In our previous reports,^{10,11} some dimeric products in the methanol-soluble portion from Japanese cedar were believed to be biphenyl-type, diphenylethane-type, stilbene-type, and phenylcoumaran-type compounds, which have bonding between guaiacyl nuclei. We also detected dimeric products in the methanol-soluble portion from Japanese beech, which may indicate guaiacyl – guaiacyl structures or guaiacyl – syringyl structures. Unfortunately, these products have not been identified by GC-MS because authentic compounds of the dimeric products are not commercially available.

MWL is regarded as one of the best samples to represent a native lignin in wood, although it is partly modified during preparation. To trace the structural change of native lignin during supercritical water treatment, the FT-IR spectra of

Table 3. Lignin-derived monomeric products in the methanol-soluble portion from Japanese cedar and Japanese beech treated in supercritical water

No.	Product	Retention time (min)	Structure ^a
1	Guaiacol	11.8	G
2	Methylguaiacol	14.8	G—C
3	Ethylguaiacol	17.2	G—C—C
4	Vinylguaiacol	18.2	G—C=C
5	Syringol	19.1	S
6	Vanillin	20.5	G—CHO
7	Eugenol	19.3	G—C—C=C
8	Propylguaiacol	19.5	G—C—C—C
9	Methylsyringol	21.4	S—C
10	Isoeugenol (<i>cis</i>)	21.8	G—C=C—C
11	Homovanillin	22.0	G—C—CHO
12	Isoeugenol (<i>trans</i>)	22.2	G—C=C—C
13	Acetoguaiacone	22.7	G—CO—C
14	Propioguaiacone ^b	23.1	G—CO—C—C
15	Ethylsyringol ^b	23.4	S—C—C
16	Guaiacylacetone ^b	23.7	G—C—CO—C
17	Vinylsyringol ^b	24.4	S—C=C
18	Propylsyringol ^b	25.6	S—C—C—C
19	2-Methoxy-4-(1-hydroxypropyl)phenol	26.5	G—C—C—C—OH
20	Allylsyringol	26.6	S—C—C=C
21	Syringaldehyde	26.7	S—CHO
22	Propenylsyringol (<i>cis</i>) ^b	26.8	S—C=C—C
23	Homovanillic acid	26.8	G—C—COOH
24	Synapylalcohol (<i>cis</i>)	27.1	S—C=C—C—OH
25	Synapylalcohol (<i>trans</i>)	27.3	S—C=C—C—OH
26	Propenylsyringol (<i>trans</i>) ^b	27.6	S—C=C—C
27	2-Methoxy-4-(prop-1-en-3-one)phenol ^c	27.9	G—CO—C=C
28	Synapylaldehyde (<i>cis</i>)	28.2	G—C=C—CHO
29	Acetosyringone	28.3	S—CO—C
30	Coniferylaldehyde (<i>trans</i>)	28.4	G—C=C—CHO
31	Propiosyringone ^b	28.6	S—CO—C—C
32	Syringylacetone ^b	29.0	S—C—CO—C
33	Ferulic acid	30.1	G—C=C—COOH
34	Synapylaldehyde (<i>trans</i>)	32.7	S—C=C—CHO

Product numbers correspond to peak labels in Fig. 2. Unless stated otherwise, products were identified from their mass spectra and by comparing their retention times with those of authentic compounds

^a G and S represent guaiacyl and syringyl nuclei, respectively

^b Products identified from literature data²¹

^c Products identified from literature data²²

the methanol-soluble portion and the MWL were recorded (Fig. 3). Aromatic nuclei were well represented in both samples as peaks at 1600, 1510, and 1425 cm⁻¹ (symbol *a*). The peaks at 1270 (symbol *g*) and 1330 cm⁻¹ (symbol *s*) represented guaiacyl nuclei and syringyl nuclei, respectively.²⁹ These peaks are typical of lignin, and suggest that aromatic nuclei are almost stable during the supercritical water treatment. However, the peaks of the methanol-soluble portion were slightly broader than those of the MWL. The peak in the methanol-soluble portion that appeared at 1030 cm⁻¹ (symbol *d*), which is assigned to aromatic C—H deformation etc.,²⁹ was smaller than that in the MWL. These features might be attributed to condensed type linkages of lignin.^{8,30}

Apart from the above-mentioned peaks, newly formed peaks were also detected in the methanol-soluble portion. The peak at 1720–1740 cm⁻¹ (symbol *n1*) indicates unconjugated ketone and carbonyl groups.²⁹ This might be due to the increase of products like **11**, **16**, **23**, and **32** (see Table 3). The peak at 980 cm⁻¹ (symbol *n2*) was attributed

to —HC=CH— out-of-plane deformation.²⁹ Considering that a dimeric product of stilbene-like structure was detected in the methanol-soluble portion,¹⁰ the *n2* peak might be caused by formation of a stilbene-like structure. In addition, the peak at around 1140 cm⁻¹ was resolved to peaks at 1125 and 1150 cm⁻¹ (symbol *n3*) in the methanol-soluble portion from Japanese cedar, and was shifted to 1125 cm⁻¹ (symbol *n4*) in that from Japanese beech. The reason for such changes is unclear. However, considering that the peak around 1140 cm⁻¹ is assigned to aromatic C—H plane deformation and secondary alcohols,²⁹ this feature may also be attributed to condensed-type linkages of lignin and modification of propyl side chains.

It is not possible to clarify the structure of the methanol-soluble portion by means of FT-IR analysis alone. However, our previous results^{8,10} were roughly consistent with these spectra. A further study, such as the isolation of a product in the methanol-soluble portion and development of application fields, is also necessary to realize effective utilization of the methanol-soluble portion.

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References

- Bobleter O (1994) Hydrothermal degradation of polymers derived from plants. *Prog Polym Sci* 19:797–841
- Saka S, Ueno T (1999) Chemical conversion of various celluloses to glucose and its derivatives in supercritical water. *Cellulose* 6:177–191
- Saka S, Konishi R (2001) Chemical conversion of biomass resources to useful chemicals and fuels by supercritical water treatment. In: Bridgwater AV (ed) *Progress in thermochemical biomass conversion*. Blackwell, Oxford, pp 1338–1348
- Ishikawa Y, Saka S (2001) Chemical conversion of cellulose as treated in supercritical methanol. *Cellulose* 8:189–195
- Tsujino J, Kawamoto H, Saka S (2004) Reactivity of lignin in supercritical methanol studied with various lignin model compounds. *Wood Sci Technol* 37:299–307
- Minami E, Saka S (2003) Comparison of the decomposition behaviors of hardwood and softwood in supercritical methanol. *J Wood Sci* 49:73–78
- Minami E, Kawamoto H, Saka S (2003) Reaction behavior of lignin in supercritical methanol as studied with lignin model compounds. *J Wood Sci* 49:158–165
- Ehara K, Saka S, Kawamoto H (2002) Characterization of the lignin-derived products from wood as treated in supercritical water. *J Wood Sci* 48:320–325
- Ehara K, Saka S (2002) A comparative study on chemical conversion of cellulose between the batch-type and flow-type systems in supercritical water. *Cellulose* 9:301–311
- Takada D, Ehara K, Saka S (2004) Gas chromatographic and mass spectrometric (GC-MS) analysis of lignin-derived products from *Cryptomeria japonica* treated in supercritical water. *J Wood Sci* 50:253–259
- Ehara K, Saka S (2004) Recent progress in transformation of lignocellulosics to fuels and chemicals by supercritical water technology. In: Saha BC, Hayashi K (eds) *Lignocellulose biodegradation*. American Chemical Society, Washington DC, in press
- Miyafuji H, Nakata T, Ehara K, Saka S (2003) Bioethanol production from supercritically-treated cellulose. In: *Proceedings of Kyoto University 21st Century COE program, The 2nd International Symposium on Bioenergy Systems, Kyoto*, pp 69–77
- Yokoyama C, Nishi K, Nakajima A, Seino K (1998) Thermolysis of organosolv lignin in supercritical water and supercritical methanol (in Japanese). *Sekiyu Gakkaishi* 41:243–250
- Saisu M, Sato T, Watanabe M, Adschiri T, Arai K (2003) Conversion of lignin with supercritical water–phenol mixture. *Energy Fuel* 17:922–928
- Funazukuri T, Wakao N, Smith M (1990) Liquefaction of lignin sulfonate with subcritical and supercritical water. *Fuel* 69:349–353
- Drews M, Barr M, Williams M (2000) A kinetic study of the SCWO of a sulfonated lignin waste stream. *Ind Eng Chem Res* 39:4784–4793
- Kratzl K, Vierhapper F (1971) Synthese von ^{14}C -kernmarkierten vanillin und bikreosolen. *Monatsh Chem* 102:425–430
- Bjorkman A (1956) Studies on finely divided wood. Part I. Extraction of lignin with neutral solvents. *Sven Papperstidn* 59:477–485
- Kusdiana D, Minami E, Ehara K, Saka S (2002) Development of the batch-type and flow-type supercritical fluid biomass conversion systems. In: *Proceedings of 12th European Conference and Technology Exhibition on Biomass for Energy, Industry and Climate Protection*, Amsterdam, pp 789–792
- Yasuda Y (1993) Sulfuric acid method. In: Usuda M, Iiyama K, Morohoshi N, Yamaguchi A (eds) *Mokuzai Kagaku Jikkensyo (Experimental Book on Wood Chemistry)*. Chugai Sangyo, Tokyo pp 156–157
- Meier D, Faix O (1992) Pyrolysis-gas chromatography-mass spectrometry. In: Dence C (ed) *Methods in lignin chemistry*. Springer, Berlin Heidelberg New York, pp 177–99
- Vane C (2003) The molecular composition of lignin in spruce decayed by white-rot fungi (*Phanerochaete chrysosporium* and *Trametes versicolor*) using pyrolysis-GC-MS and thermochemical analysis with tetramethylammonium hydroxide. *Int Biodeter Biodegr* 51:67–75
- Bobleter O, Concin R (1979) Degradation of poplar lignin by hydrothermal treatment. *Cell Chem Technol* 13:583–593
- Concin R, Burtscher E, Bobleter O (1981) The molecular weight distribution of hydrothermally degraded lignin. *Holzforschung* 35:279–282
- Concin R, Burtscher P, Burtscher E, Bobleter O (1983) Separation and identification of monomeric lignin degradation products by GLC-mass spectrometry. *Int J Mass Spectrom Ion Phys* 48:63–66
- Kallury R, Tidwell T, Agblevor F, Boocock D, Holysh M (1987) Rapid hydrothermolysis of poplar wood: comparison of sapwood, hardwood, bark and isolated lignin. *J Wood Chem Technol* 7:353–371
- Agblevor F, Boocock D (1989) The origins of phenol produced in the rapid hydrothermolysis and alkaline hydrolysis of hybrid poplar lignins. *J Wood Chem Technol* 9:167–188
- Sato T, Sekiguchi G, Saisu M, Watanabe M, Adschiri T, Arai K (2002) Dealkylation and rearrangement kinetics of 2-isopropylphenol in supercritical water. *Ind Eng Chem Res* 41:3124–3130
- Faix O (1991) Classification of lignins from different botanical origins by FT-IR spectroscopy. *Holzforschung* 45 Suppl:21–27
- Winston MH, Chen C-L, Gratzl JS, Goldstein IS (1986) Characterization of the lignin residue from hydrolysis of sweetgum wood with superconcentrated hydrochloric acid. *Holzforschung* 40 Suppl:45–50