Electronic Supplementary Information

Organosolv processing of Sitka Spruce sawdust: Large scale preparation of native-like lignin and lignin^{ox} for valorisation

Daniel J. Davidson[a+], Geraud N. Sansom[a+], Daniel M. Miles-Barrett[a], David B. Cordes[a], Alexandra M.Z. Slawin[a], James R. D. Montgomery[a], Tomas Lebl[a], Ann Connor[b], Andrew M. Danby[c], Mark J. Gronnow[c], Neil J. Parry[b*], Sarah L. Hosking[b*], David S. Grainger[b*], Nicholas J. Westwood[a*]

+ these authors contributed equally

* co-corresponding authors

[a] School of Chemistry and Biomedical Sciences Research Complex, University of St. Andrews and EaStCHEM, St Andrews, Fife, Scotland, KY16 9ST, UK. [b] Unilever Research and Development, Port Sunlight Laboratory, Quarry Road East, Bebington, Wirral, UK. [c] Biorenewables Development Centre, University of York, Unit 1 Hassacarr Close, Chessingham Park, Dunnington, York, YO19 5SN, UK.

General Considerations

Reagents and Solvents

Commercially available compounds were purchased and used as received from chemical suppliers (Sigma-Aldrich, Merck, Fisher Scientific, ThermoFisher Scientific, Fluorochem, BLD Pharma, TCI, Strem) unless otherwise stated.

Dry solvents were obtained from a MBraun SPS-800 solvent dispenser system.

Biomasses were dried to constant weight then milled prior to pretreatments.

Equipment

Solution-state NMR spectra were obtained using Bruker AV-II 400, Bruker Neo 400 fitted with BFF-H-D iProbe, Bruker AV-III 500 fitted with CryoProbe Prodigy BBO, Bruker AV-III HD 500 BBFO+, and Bruker AV-III-HD 700 fitted with CryProbe Prodigy TCI. In some cases ^{13}C NMR signals are reported to 2 decimal places to distinguish between nearly overlapping signals.

FT-IR spectra were obtained using a Shimadzu IRAffinity 1S IR Spectrometer as ATR.

Mass spectrometry data were acquired through the University of St Andrews School of Chemistry mass spectrometry service using a Thermo Exactive Orbitrap mass spectrometer using ESI ionisation in both positive and negative mode.

Melting points were measured using a Stuart SMP10 Digital Melting Point Apparatus with a heating rate of 2 °C/min with an accuracy of ± 1.0 °C at 20 °C and ± 2.5 °C at 300 °C.

UV-Vis spectra were recorded using a Jasco V-650 UV-Vis double beam spectrophotometer with a deuterium lamp light source between 190-350 nm and a halogen lamp light source between 330-900 nm with a scanning speed of 400 nm/min.

Lignin NMR Sample Preparation

Lignin (60.0 ± 0.1 mg) was dissolved in DMSO-d_6 (700 µL) and sonicated for 10 minutes to ensure complete dissolution. Samples were analysed using ^{1}H, HSQC, HMBC and ^{31}P NMR (where appropriate).

General Procedures

General Procedure A – Quantification of Lignin using Cysteine-Assisted Sulfuric Acid (CASA) Method[S1]

A stock solution of cysteine in 12M sulfuric acid (0.1 g/mL) was prepared. Dried SiS sawdust (~ 7 mg) was stirred in the stock solution (1 mL) for 1 – 2 hours until completely dissolved. The sample was diluted with deionised water to 100 mL and the UV-Vis spectrum of the diluted solution recorded between 230 nm to 400 nm.

General Procedure B – Standard Pressure Bronze Standard Organosolv Pretreatment

SiS sawdust was suspended in the alcohol under test (9.5 mL/g) and heated to reflux. 4M HCl (0.5 mL/g) was added when at reflux and then the suspension was stirred vigorously for 6 hours. The suspension was then cooled to room temperature, filtered, washed through with acetone (2 x 2.5 mL/g), neutralised with sat. aq. sodium bicarbonate and concentrated under reduced pressure. The resulting solid was dissolved in the minimum volume of 9:1 acetone/water, precipitated into 0.1M HCl (10 v/v eq.), filtered, washed with excess water until neutral and dried under vacuum at 50 °C for 24 hours to afford bronze standard organosolv lignin.

General Procedure C – Pressurised Bronze Standard Organosolv Pretreatment

SiS sawdust was suspended in 19:1 alcohol/4M HCl (10 mL/g) inside a sealed pressure tube. The vessel was sealed and heated to 20 °C above the boiling point of the alcohol for 6 hours with vigorous stirring. The suspension was cooled to room temperature, filtered, washed through with acetone (2 x 2.5 mL/g), neutralised with sat. aq. sodium bicarbonate and concentrated under reduced pressure. The resulting solid was dissolved in the minimum volume of 9:1 acetone/water, precipitated into 0.1M HCl (10 v/v eq.), filtered, washed with excess water until neutral and dried under vacuum at 50 °C for 24 hours to afford bronze standard organosolv lignin.

General Procedure D – Silver and Gold Standard Butanosolv

SiS sawdust was treated as in General Procedure C to afford bronze standard butanosolv lignin which was dissolved in the minimum volume of 9:1 acetone/methanol, precipitated into 1:1 hexane/ether (10 v.v eq.) and filtered to give silver standard lignin. The residue was suspended in 0.1M NaOH (10 mL/g), heated to 50 °C for 18 hours then precipitated into 0.1M HCl (10 v/v eq.). The residue was dissolved in the minimum volume of 9:1 DCM/methanol and dry-loaded onto silica gel (5 g) then purified by flash column chromatography on silica gel (40 g) eluting with 0-100% DCM/hexane followed by 0-10% methanol/DCM and a 10% methanol/acetone flush. The combined lignin containing fractions were concentrated under reduced pressure and the resulting solid was dried under vacuum at 50 °C for 24 hours to afford gold standard butanosolv lignin.

General Procedure E – Phosphitylation of Lignin for Quantitative ^{31}P NMR Analysis[S2-4]

Lignin (~30 mg) was dissolved in 1:1.6 $CDCl_3$/pyridine (500 µL), containing cyclohexanol (10 µL) as an internal standard, and sonicated for 10 minutes to ensure complete dissolution. 2-Chloro-4,4,5,5-tetramethyl-1,3,2-dioxaphospholane (50 µL) was added and the combined sample diluted with $CDCl_3$ (500 µL). The quantitative ^{31}P NMR experiment was run within 6 hours of sample preparation. The internal standard integral was set to 1.00 and referenced to 145.15 ppm.[S2-4]

General Procedure F – Lignin Acetylation for Gel-Permeation Chromatogprahy (GPC)/Size Exclusion Chromatography (SEC)

The under analysis lignin (~7 mg) was dissolved in pyridine (0.5 mL). Acetic anhydride added (0.5 mL) and the reaction was stirred at room temperature for 18 hours. After concentrated under reduced pressure the resulting solid was dried azeotropically using toluene (3 x 15 mL) and then evaporated from ethanol (3 x 15 mL) then DCM (3 x 15 mL). Acetylated lignin was prepared for analysis by dissolving in HPLC-grade THF (1 mL) prior to injection.

General Procedure G – Large scale reversion of bronze butanosolv SiS Lignin

Larger scale reversions were performed at the Biorenewables Development Centre using a 2 L Hastelloy high pressure reactor (Series 4530, Parr Instruments Company, Moline, IL). In a typical reaction, 60 g of lignin were added to the reactor followed by 222 ml of water, 111 ml of 1 M HCl(aq) and finally 667 ml of dioxane. The reactor was sealed and then heated, with stirring, to 105 °C. Heating was continued for 2.5 hours and an autogenous pressure of *ca*. 0.2 barg was observed. The reactor was then cooled to room temperature, opened and the contents slowly added to 0.1 M HCl(aq) (10 L). The resulting precipitate was filtered off, washed with water (5 L) and then dried in a vacuum oven at 40 °C for two to three days.

General Procedure H - Lignin oxidation

Reverted lignin and DDQ (10-25% wt eq.) were added to a 100 mL Asynt high pressure reactor with a PTFE inner tube. At larger scales, a 2 L Hastelloy high pressure reactor (Series 4530, Parr Instruments Company, Moline, IL) was used instead. The solvent mixture was added (1,4-dioxane:DME, 2:3). Just before closing the reaction vessel, ^{t}BuONO (10-25% wt eq.) was added as a liquid via a micropipette. The reaction vessel was sealed and pressurised up to 10 atm. of compressed air and heated at 85 °C overnight. To end the reaction, the heating was removed and once the vessel had cooled down the excess pressure was released. The resulting lignin was isolated by precipitation into 10 volumes of Et_2O and filteration.

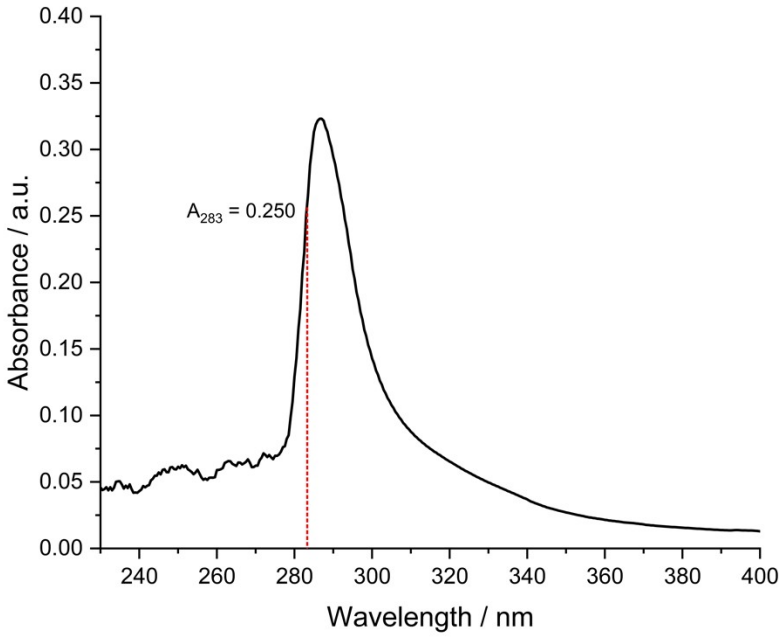
General Procedure I - Furan aldehyde synthesis

Step 1: To a stirring solution of resinol (1.00 eq.) in 1,4-dioxane (*C* = 0.02M) was added DDQ (5.00 eq.). The reaction was heated to 80 °C for 3 hours. Upon completion, the reaction was cooled to room temperature (DDQ-H_2 precipitated better at lower temperatures) and filtered through Celite. The filtrate was diluted with EtOAc, washed with $Na_2S_2O_3$ (freshly prepared sat. aqueous solution), $NaHCO_3$ (freshly prepared sat. aqueous solution), brine, dried over $MgSO_4$ and concentrated *in vacuo*.

Step 2: To the crude mixture of furan-alcohol and furan-aldehyde in DCM (*C* = 0.02M) was added Dess-Martin periodinane (DMP, 1.00 eq.). The reaction was stirred at room temperature

for 1 hour. The reaction was quenched with $Na_2S_2O_3$: $NaHCO_3$ (1:1) and extracted with DCM (3 times). The combined organic extracts were washed with brine, dried over $MgSO_4$ and concentrated *in vacuo.*

Results from CASA analysis



$$\text{Lignin Content} = \frac{\text{Abs}_{283} \times V}{\varepsilon \times m_s \times L} \times 100 = 21.3 \text{ wt. \% of lignin in SiS}$$

Where Abs_{283} = absorbance at 283 nm / a.u. (0.250 a.u.)

V = total volume of diluted solution / L (0.1 L)

ε = extinction coefficient (17.25 g^{-1} L cm^{-1} for softwoods)[S1]

m_s = mass of biomass sample used / g (0.0068 g)

L = path length / cm (1 cm)

Figure S1. UV-Vis spectrum of Sitka spruce biomass dissolved according to CASA method[S1] between 230 nm to 400 nm.

Quantitative ^{31}P NMR analysis of phosphitylated lignins[S2-4]

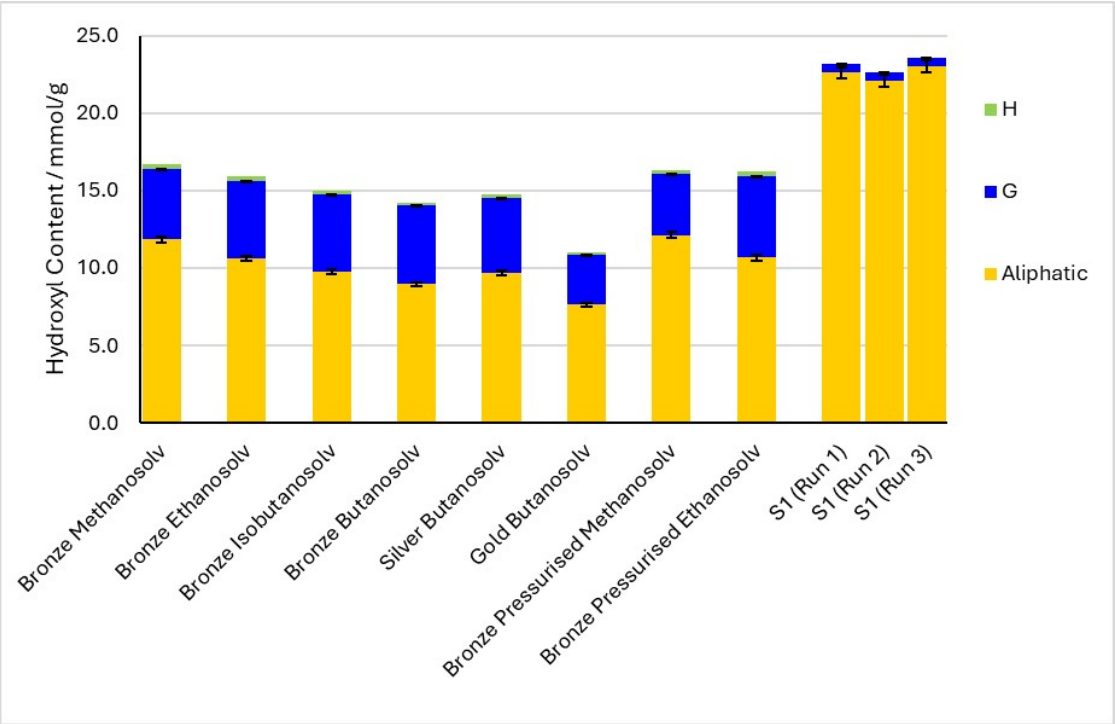


Figure S2. Hydroxyl content of SiS organosolv lignins calculated using quantitative ^{31}P NMR analysis after lignin phosphitylation. Phosphitylation of model all G β-O-4 oligomer **S1** was carried out in triplicate and used to calculate an error (standard deviation) of 1.7% in aliphatic hydroxyl content and 0.3% in G phenolic content. No H content was present in **S1** and so no error was calculated for H content in the lignin. For experimental protocol see General Procedure E.

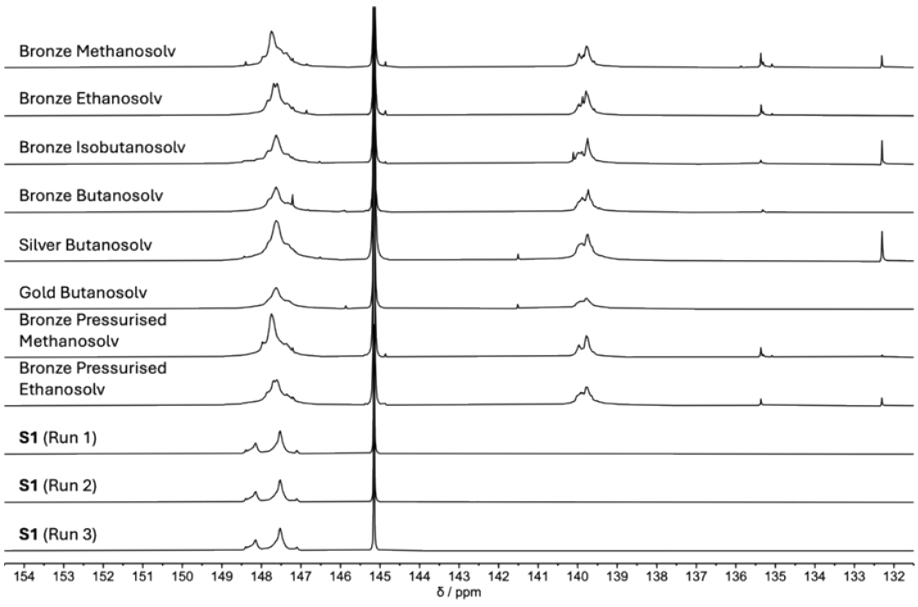


Figure S3. Quantitative ^{31}P NMR spectra of SiS organosolv lignins after phosphitylation.

Table S1. Mass of sample used and integrals of regions used to calculated hydroxyl content of SiS organosolv lignins after phosphitylation (relative to internal standard integral set to 1.00).

Lignin	Mass of Sample / mg	Relative Integral				
		Aliphatic	S	G	H	COOH
Bronze Methanosolv	30.1	1.07	0.03	0.41	0.03	0.07
Bronze Ethanosolv	30.1	0.96	0.02	0.45	0.03	0.06
Bronze Isobutanosolv	30.1	0.88	0.03	0.45	0.02	0.03
Bronze Butanosolv	30.2	0.81	0.04	0.46	0.02	0.02
Silver Butanosolv	29.9	0.87	0.05	0.43	0.02	0.00
Gold Butanosolv	30.2	0.69	0.03	0.29	0.02	0.01
Bronze Pressurised Methanosolv	29.7	1.08	0.03	0.35	0.02	0.04
Bronze Pressurised Ethanosolv	30.0	0.96	0.07	0.47	0.03	0.04
S1 (Run 1)	30.1	2.04	0.00	0.05	0.00	0.00
S1 (Run 2)	30.2	2.01	0.00	0.05	0.00	0.00
S1 (Run 3)	30.0	2.01	0.00	0.04	0.00	0.00

Table S2. Hydroxyl group content of SiS organosolv lignins calculated from quantitative ^{31}P NMR after phosphitylation.

Lignin	Hydroxyl Group Content / mmol/g[a]						
	Aliphatic	S	G	H	COOH	Total Phenolic	Total
Bronze Methanosolv	11.85	0.33	4.54	0.33	0.78	5.20	17.83
Bronze Ethanosolv	10.63	0.22	4.98	0.33	0.66	5.54	16.83
Bronze Isobutanosolv	9.75	0.33	4.98	0.22	0.33	5.54	15.61
Bronze Butanosolv	8.94	0.44	5.08	0.22	0.22	5.74	14.90
Silver Butanosolv	9.70	0.56	4.79	0.22	0.00	5.57	15.27
Gold Butanosolv	7.62	0.33	3.20	0.22	0.11	3.75	11.48
Bronze Pressurised Methanosolv[b]	12.12	0.34	3.93	0.22	0.45	4.49	17.06
Bronze Pressurised Ethanosolv[b]	10.67	0.78	5.22	0.33	0.44	6.33	17.44
S1 (Run 1)	22.59	0.00	0.55	0.00	0.00	0.55	23.15
S1 (Run 2)	22.08	0.00	0.55	0.00	0.00	0.55	22.63
S1 (Run 3)	23.00	0.00	0.56	0.00	0.00	0.56	23.56

[a]Hydroxyl content is measured in mmol/g using the ^{31}P phosphitylation method. This means that for theoretically analogous alkosolv lignins that differ only in the alcohol used (same average chain length and number of alcohol modified β-O-4 units per 100 C9 units) then a change should still be seen in the numerical values determined. As the average molar mass of an alkoxylated β-O-4 linkage decreases (for example when going from a butanosolv lignin to a methanosolv lignin), the hydroxyl content per gram should increase even for an analogous lignin (same average chain length and number of alcohol modified β-O-4 units per 100 C9 units, Figure S4). This is in agreement with the observed trend in this study. Methanosolv lignin has apparently greater hydroxyl content than ethanosolv lignin which in turn had apparently greater hydroxyl content than butanosolv and isobutanosolv lignins. Clearly the actual situation is more complicated than this; [b]Pressurised methanosolv and ethanosolv lignins had an aliphatic hydroxyl content within 0.3 mmol/g of the equivalent standard pressure lignins. This confirmed that the main benefit of pressurising the system for the organosolv was on the increased yield of the isolated lignin.

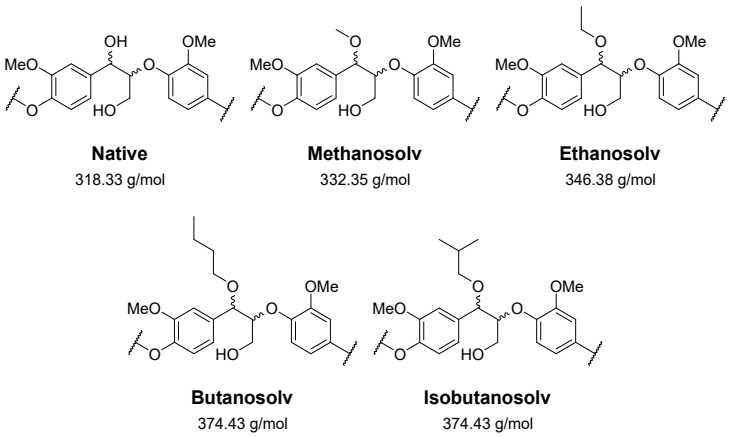
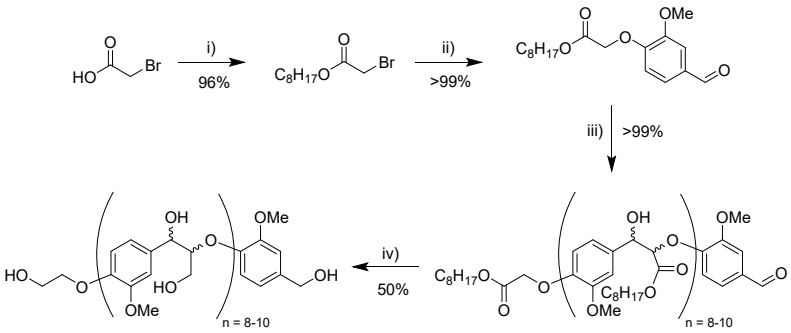


Figure S4. Calculated MWs of native and alkylated β-O-4 units in organosolv lignins.

Synthesis of β-O-4 Model Oligomer S1

β-O-4 model oligomer **S1** was prepared according to a literature procedure[S5] with some modifications (Scheme S1).



Scheme S1. Synthesis of β-O-4 model oligomer **S1**.[S5] i) *n*-octanol (1.0 eq.), cat. *p*TSA.H_2O, cyclohexane, Dean-Stark reflux, 16 hours; ii) vanillin (1.0 eq.), K_2CO_3 (2.0 eq.) acetone, reflux, 16 hours; iii) LDA (1.5 eq.), dry THF, -78 °C to room temperature, 3 hours; iv) $NaBH_4$ (5 eq.), methanol (15 eq.), ethanol, 50 °C, 3 hours. The previously reported literature yield of **S1** was 33%[S5] as a single component after purification by precipitation into organic solvent. Repeating the literature procedure[S5] on a 20 g scale of the octyl ester monomer gave a yield of 50% of **S1** prior to dissolution for the organic precipitation. When dissolution in 9:1 acetone/ethanol was attempted, a soluble component (4%) and an insoluble residue component (45%) were formed. The two fractions were found to have different molecular weights by DOSY NMR analysis. The molecular weights of the fractions were calculated to be 2350 ± 900 Da and 4320 ± 900 Da for the soluble and insoluble fractions, respectively. A second large scale batch of **S1** was prepared using 25 g of the octyl ester monomer, this time without fractionation by precipitation. This showed a further improvement in the yield of **S1** (8.22 g, 57%). GPC analysis of this larger second batch of **S1** gave calculated molecular weights of M_n = 1896 Da and M_w = 2967 Da (PDI = 1.565) which was more closely representative of the molecular weights of the lignins obtained in this work (Table 1 in the main text) and so this batch was used as the reference material in this work. Reasons for the variability across batches remain elusive.

HSQC NMR analysis of lignin from Pretreatments

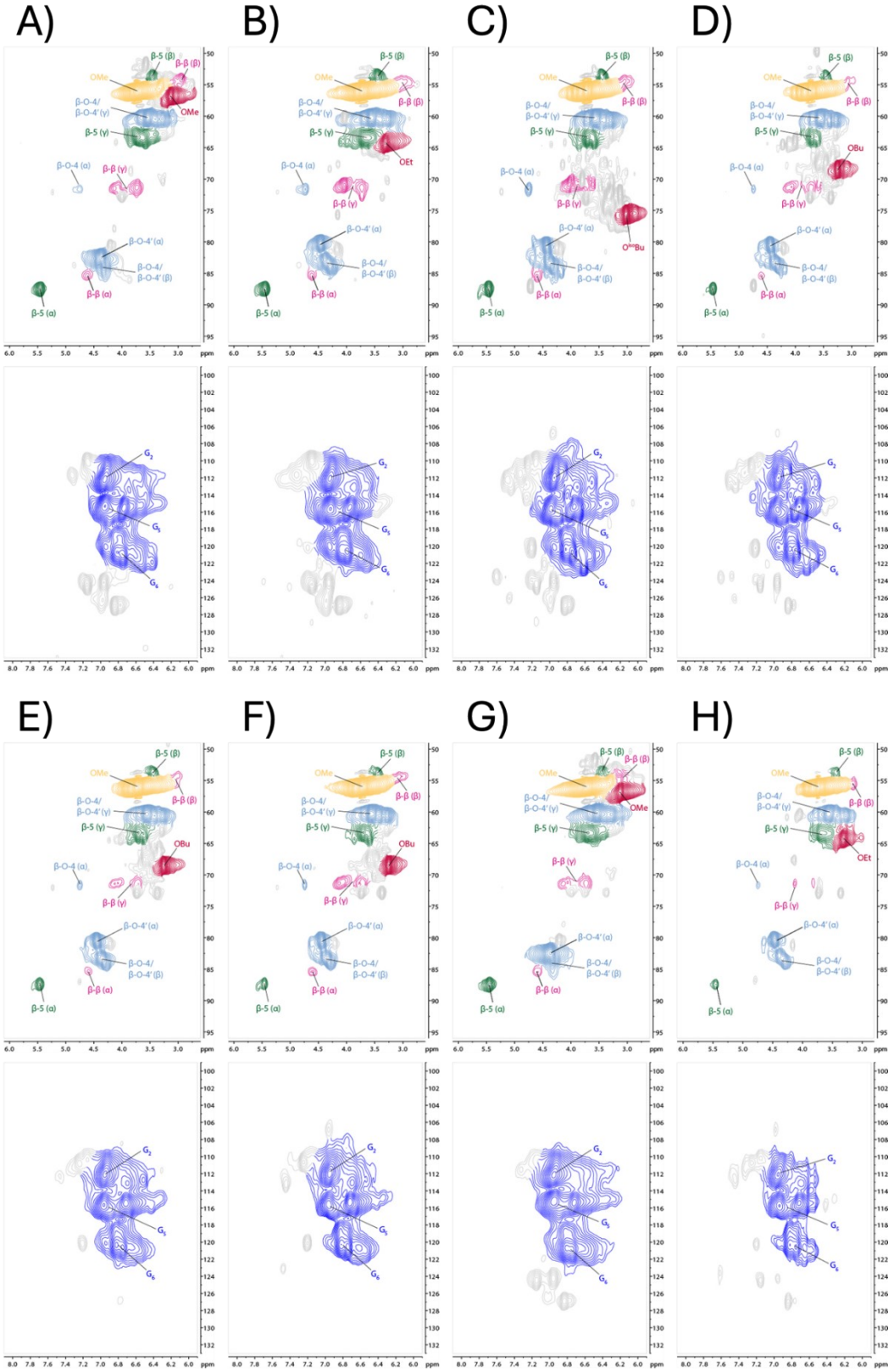
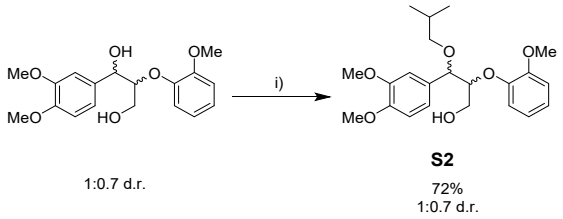


Figure S5. HSQC NMR (700 MHz, DMSO-d6) analysis of the linkage region (top) and aromatic region (bottom) of lignins: A) bronze methanosolv; B) bronze ethanosolv; C) bronze isobutanosolv; D) bronze butanosolv; E) silver butanosolv; F) gold butanosolv; G) bronze pressurised methanosolv; H) bronze pressurised ethanosolv.

The linkage content determined semi-quantitatively using HSQC NMR analysis is a useful value for comparing the relative abundance of the different linkages between lignins. However, the value for the β-O-4 Model Oligomer **S1** was only 60 β-O-4 linkages per 100 C9 units (data not shown) rather than the expected 100 units (as no other linkages should be present). This highlights the limitations and inaccuracy of this method and that additional analytical methods are required for a more robust comparison between the protocols.

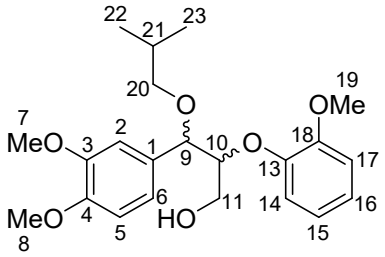
Assignment of Isobutanosolv Lignin using Model Compound S2

Isobutanosolv pretreatment has not been reported previously in the literature, to the best of our knowledge. Novel model compound **S2** was synthesised (Scheme S2) and compared to the SiS isobutanosolv lignin (Figure S6) to aid in assignment of isobutoxylated β-O-4 linkages. Comparison of the HSQC NMR spectrum of **S2** (Figure S6A) with that of the SiS isobutanosolv lignin (Figure S6B) confirmed that isobutoxylation had taken place during the pretreatment to afford the α-isobutoxylated lignin.



Scheme S2. Synthesis of isobutanosolv model dimer **S2**. i) Isobutanol, 4M HCl, reflux, 20 minutes.

Synthesis of S2



Non-phenolic β-O-4 dimer[S6] (0.55 g, 1.63 mmol, *major* : *minor diastereomer ratio* 1:0.7) was dissolved in isobutanol (5.5 mL) and heated to reflux. 4M aqueous HCl (0.5 mL) was then added and the reaction was heated at reflux for 20 minutes. The resulting mixture was cooled and quenched with sat. aq. sodium bicarbonate (15 mL), extracted with ethyl acetate (3 x 15 mL). The combined organic extracts were washed with brine (1 x 15 mL), dried over $MgSO_4$ and concentrated under reduced pressure. Purification by column chromatography, eluting with ethyl acetate/hexane (0% to 50%) gave the diastereomeric mixture **S2** (0.46 g, 72%, *major*/*minor* 1:0.7) as a viscous yellow oil.

IR (ATR) ν_{max} 2954, 1591, 1500, 1456, 1251, 1024, 744 cm^{-1}.

Major:

^{1}H NMR (500 MHz, DMSO-d_6) δ 7.02 – 6.97 (m, 2H, H2, H5), 6.94 – 6.89 (m, 1H, H14), 6.90 – 6.86 (m, 2H, H16, H17), 6.86 – 6.77 (m, 2H, H6, H15), 4.69 (t, J = 5.6 Hz, 1H, OH12), 4.48 – 4.44 (m, 1H, H9), 4.44 – 4.40 (m, 1H, H10), 3.72 (s, 3H, CH_3), 3.70 (s, 3H, CH_3), 3.68 (s, 3H, 3 x H7), 3.63 – 3.51 (m, 2H, 2 x H11), 3.08 – 2.99 (m, 2H, 2 x H20), 1.79 – 1.67 (m, 1H, H21), 0.82 (d, J = 5.5 Hz, 3H, $CHCH_3$, 3 x H22 or 3 x H23), 0.81 (d, J = 5.6 Hz, 3H, $CHCH_3$, 3 x H22 or 3 x H23).
^{13}C NMR (126 MHz, DMSO-d_6) δ 149.8 (C3), 148.3 (ArC), 148.1 (ArC), 148.0 (ArC), 131.1 (C1), 121.1 (C6), 120.6 (C15), 120.2 (C16), 116.0 (C5), 112.6 (C14), 111.4 (C2), 111.0 (C17), 82.5 (C10), 80.1 (C9), 75.1 (C20), 59.9 (C11), 55.6 (C7), 55.4 (C19), 55.2 (C8), 28.1 (C21), 19.3 (C22 or C23), 19.2 (C22 or C23).

Minor:
^{1}H NMR (500 MHz, DMSO-d_6) δ 7.02 – 6.97 (m, 2H, H2, H5), 6.94 – 6.89 (m, 1H, H14), 6.90 – 6.86 (m, 2H, H16, H17), 6.86 – 6.77 (m, 2H, H6, H15), 4.66 (t, J = 5.5 Hz, 1H, OH12), 4.48 – 4.44 (m, 1H, H9), 4.35 – 4.30 (m, 1H, H10), 3.75 (s, 3H, H7), 3.73 (s, 3H, CH_3), 3.72 (s, 3H, CH_3), 3.63 – 3.51 (m, 1H, 1 x H11a), 3.33-3.28 (m, 1H, 1 x H11b), 3.08 – 2.99 (m, 2H, 2 x H20), 1.79 – 1.67 (m, 1H, H21), 0.79 (d, J = 6.8 Hz, 3H, 3 x H22 or 3 x H23), 0.78 (d, J = 6.7 Hz, 3H, 3 x H22 or 3 x H23).
^{13}C NMR (126 MHz, DMSO-d_6) δ 149.6 (C3), 148.8 (ArC), 148.4 (ArC), 148.2 (ArC), 131.5 (C1), 120.9 (C6), 120.6 (C15), 119.7 (C16), 115.5 (C5), 112.6 (C14), 111.2 (C2), 110.9 (C17), 83.2 (C10), 80.4 (C9), 75.4 (C20), 60.2 (C11), 55.6 (C7), 55.4 (C19), 55.3 (C8), 28.1 (C21), 19.3 (C22 or C23), 19.2 (C22 or C23).

HRMS (ESI) calculated for $C_{22}H_{30}O_6Na$ $[M+Na]^+$ 413.1940; found 413.1928.

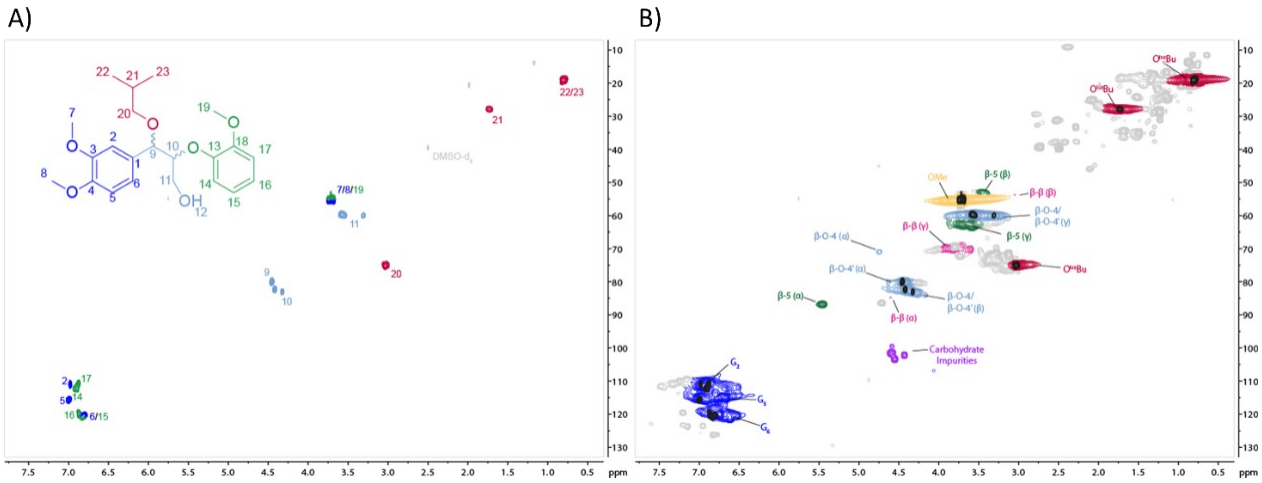


Figure S6. HSQC NMR (500 MHz, DMSO-d_6) analysis of A) isobutanosolv dimer model **S2** with colour-coded assignment of cross-peaks; and B) SiS isobutanosolv lignin overlaid with **S2** (black).

GPC Analysis of Lignins

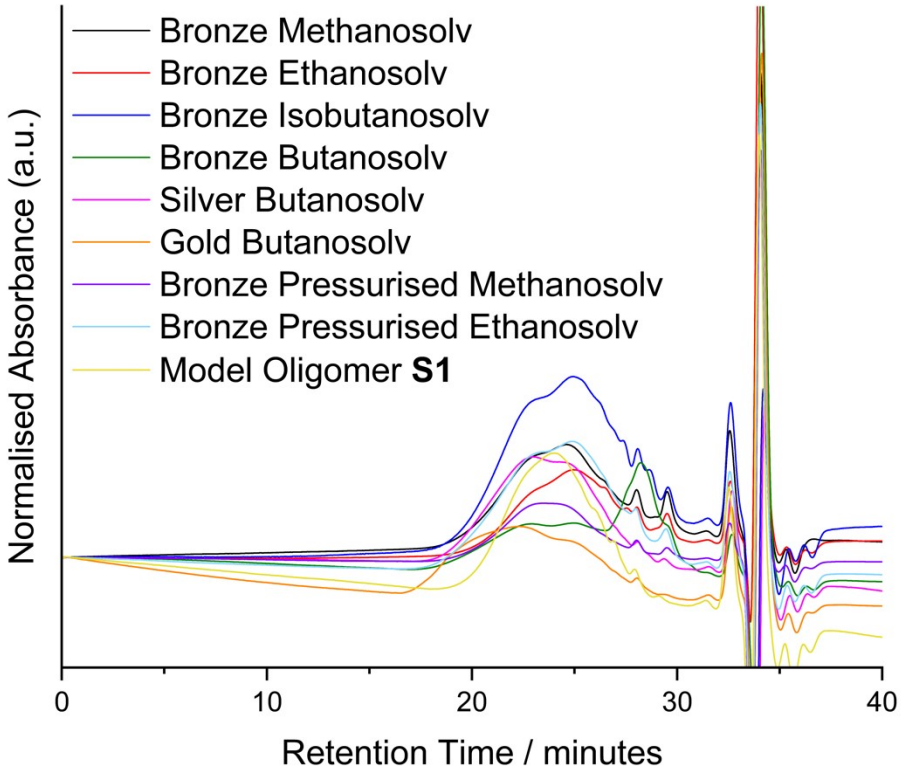


Figure S7. GPC chromatograms (Refractive Index) of the alkosolv lignins used in this study. Gel Permeation Chromatography (GPC)/Size Exclusion Chromatography (SEC) data were obtained using a Shimadzu Prominence HPLC System equipped with: CMB-20A Communications Bus Module; DGU-20A$_{SR}$ Degassing Unit; LC-20AD Pump; SIL-20A HT Auto Sampler; CTO-20A Column Oven; RID-10A Refractive Index Detector. Samples were passed through the system at 1 mL/min with THF as the eluent. The following columns were used and connected in series in this order: Phenogel 5µ Guard Column 50x7.8 mm; Phenogel 5µ 10^3 Å New Column 300x7.8 mm; Phenogel 5µ 500 Å New Column 300x7.8 mm; Phenogel 5µ Guard Column 50x7.8 mm; Phenogel 5µ 50 Å New Column 300x7.8 mm. The column oven was set to 35 °C. See General Procedure F for sample preparation.

Lignin Value Factor Calculation

The LVF values are shown in Table 2 in the main manuscript.

The initially derived equation was as follows:

$$\frac{\left(\frac{Mass\ of\ biomass\ (g)}{Volume\ of\ solvent\ (L)}\right) x \left(\frac{Mass\ of\ isolated\ lignin\ (g)}{Mass\ of\ biomass\ (g)}\right) x\ Hydroxyl\ Content\ (mmol/g)}{Cost\ of\ Solvent\ (USD/L)} = \text{Lignin Value Factor}$$

Which can be simplified be to give

$$\frac{\left(\frac{Mass\ of\ biomass\ (g)}{Volume\ of\ solvent\ (L)}\right) x \left(\frac{Mass\ of\ isolated\ lignin\ (g)}{Mass\ of\ biomass\ (g)}\right) x\ Hydroxyl\ Content\ (mmol/g)}{Cost\ of\ Solvent\ (USD/L)} = \text{LVF}$$

and therefore

Equation 1

$$\frac{\left(\frac{m_{lignin}}{V_{solvent}}\right) x\ HC}{Cost\ of\ Solvent} = \text{Lignin Value Factor / mmol/USD}$$

Where m_{lignin} = mass of isolated lignin / g

$V_{solvent}$ = volume of alcohol solvent / L

HC = aliphatic hydroxyl content (from quantitative ^{31}P NMR) / mmol/g

Cost of Solvent = Cost of solvent / USD/L (converted from Yuan/tonne)

Table S3. Densities of alcohol solvents used in SiS organosolv study.

Solvent	Density / g/cm^3
Methanol	0.792
Ethanol	0.789
Isobutanol	0.803
Butanol	0.810

Table S4. Cost of alcohol solvents used in SiS organosolv study in Yuan/tonne (converted to USD/L for LVF calculation) obtained from ECHEMI[1]

Solvent	Solvent Cost / Yuan/tonne	
	28th March 2024	28th March 2025
Methanol	2583.33	2654.17
Ethanol	6150.00	5250.00
Isobutanol	7600.00	7350.00
Butanol	7816.67	6633.33

Table S5. Starting material and lignin product quantities for SiS organosolv study.

Pretreatment	Mass of Biomass / g	Volume of Solvent / mL	Mass of Lignin Isolated / g
Bronze Methanosolv	50[a]	475	1.54
Bronze Ethanosolv	50[a]	475	1.86
Bronze Isobutanosolv	41[a]	380	3.19
Bronze Butanosolv	31	285	2.95
Bronze Pressurised Methanosolv	20[b]	190	1.86
Bronze Pressurised Ethanosolv	20[b]	190	1.58

[a]Larger scale experiments were carried out to provide sufficient material for analysis and to compensate for the expected lower yield compared to bronze butanosolv; [b]smaller scale experiments were carried out due to the limited size of the pressure vessel.

Reversion of Sitka spruce bronze butanosolv lignin

Table S6. Summary of reversion experiments.

Entry	Scale / g	Product / g	Yield (wt. %)	Native αOH-β-O-4 content (per 100 C9 units)	Remaining αBuO-β-O-4 content (per 100 C9 units)	Reversion (%, native αOH-β-O-4/ total α(OH+BuO)-β-O-4
1	-	-	NA[a]	3.9	40.0	8.9
2[a]	0.41	0.29	72	27.2	9.0	75.1
3[b]	0.50	0.21	42	5.2	44.0	10.5
4[c]	0.51	0.25	48	28.5	9.7	74.5
5[d]	5.03	2.85	57	26.0	19.0	57.8
6[e]	8.01	5.17	65	21.7	13.3	61.9
7[f]	6.00	3.79	63	22.4	17.3	56.4
8[g]	6.00	3.49	58	22.0	12.6	63.7
9[h]	40.0	25.9	65	22.7	9.4	70.7
10[h]	60.0	37.92	63	23.1	10.0	69.9
11[h]	60.0	38.4	64	24.3	9.6	71.7
12[h]	60.0	35.7	60	25.2	11.3	69.0
13[h]	60.0	36.3	61	22.0	8.6	71.8

Reaction conditions: [a] water:1M HCl(aq):1,4-dioxane (2.4 mL:1.0 mL:6.7 mL), sealed tube, 100 °C, 6 hours; [b] water:1M HCl(aq):1,4-dioxane (3.0 mL:1.3 mL:8.4 mL), sealed tube, RT, 3 days (with 5 wt% $BiOTf_3$); [c] water:1M HCl(aq):1,4-dioxane (3.0 mL:1.3 mL:8.4 mL), sealed tube, 100 °C, 2.5 hours; [d] water:1M HCl(aq):1,4-dioxane (27 mL:14 mL:84 mL), sealed tube, 100 °C, 2.5 hours; [e] water:1M HCl(aq):1,4-dioxane (44.4 mL:22.2 mL:133.3 mL), 350 mL sealed vessel, 105 °C, 2.5 hours; [f] water:1M HCl(aq):1,4-dioxane (33.3 mL:16.7 mL:100 mL), 350 mL sealed vessel, 105 °C, 2.5 hours; [g] water:1M HCl(aq):1,4-dioxane (44.4 mL:22.2 mL:133.3 mL), 350 mL sealed vessel, 105 °C, 2.5 hours; [h] see General Procedure G.

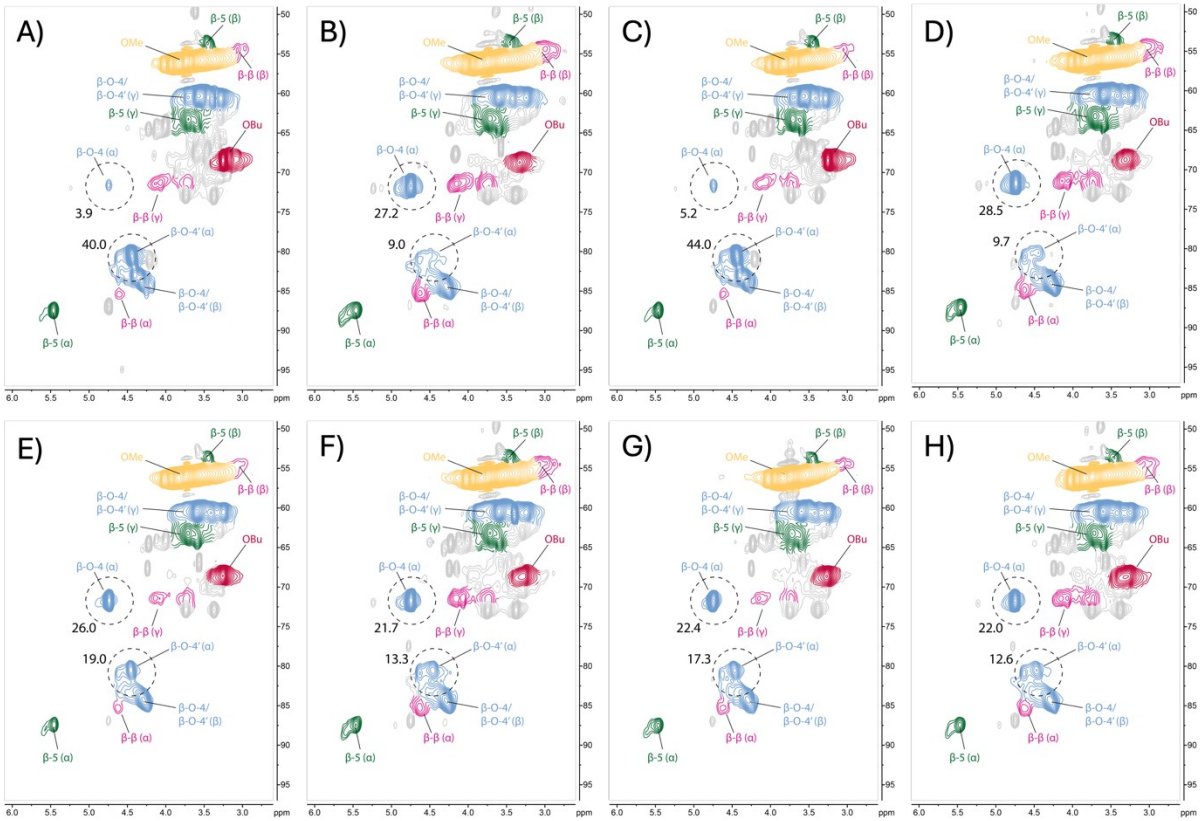


Figure S8. HSQC NMR (700 MHz, DMSO-d_6) analysis of A) starting bronze butanosolv lignin (Table S7, Entry 1) with colour-coding of cross peaks (blue = β-O-4) and with highlighting (dashed circles) of the regions corresponding to αBuO-β-O-4 and αOH-β-O-4 with a low abundance of signals corresponding to native β-O-4 linkages (αOH-β-O-4); lignins isolated from various attempts at the reversion process corresponding to entries in Table S7 - B) Entry 2; C) Entry 3; D) Entry 4; E) Entry 5; F) Entry 6; G) Entry 7; H) Entry 8. Values of the relative integrations are taken when the region corresponding to the 3 aromatic signals of the G unit of the lignins is set to 300.0.

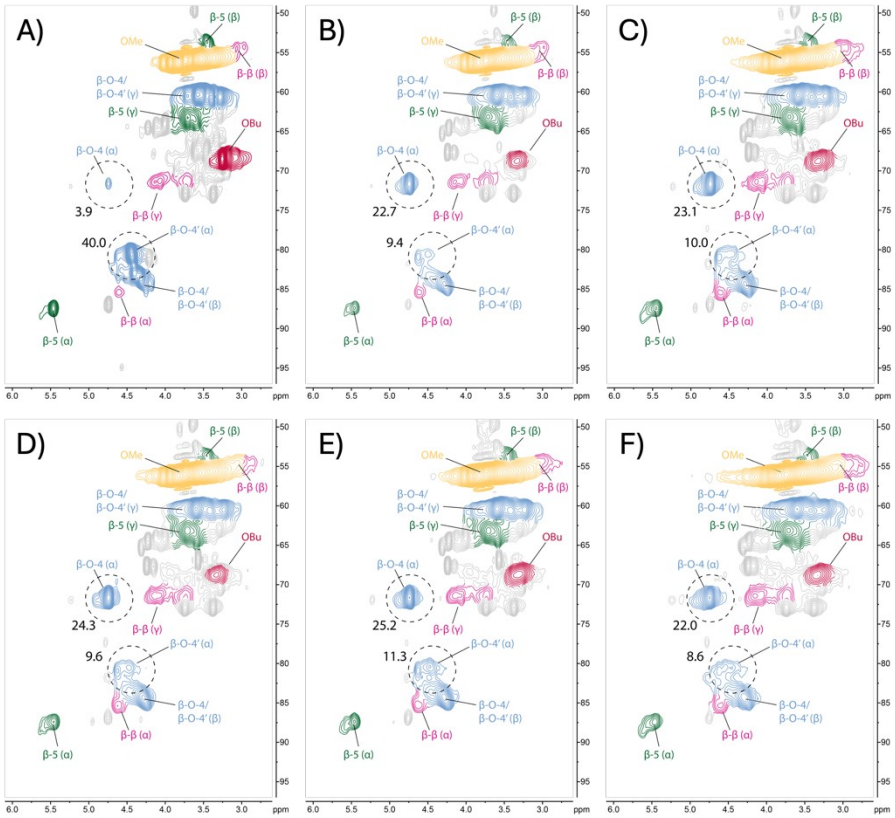


Figure S9. HSQC NMR (700 MHz, DMSO-d_6) analysis of A) starting bronze butanosolv lignin (Table S7, Entry 1) with colour-coding of cross peaks (blue = β-O-4) and with highlighting (dashed circles) of the regions corresponding to αBuO-β-O-4 and αOH-β-O-4 with a low abundance of signals corresponding to native β-O-4 linkages (αOH-β-O-4); B) lignin isolated from 40 g scale reversion carried out at Biorenewables Development Centre (Table S7, Entry 9); and lignins isolated from repeats of the large scale reversion process at 60 g scale to assess the reproducibility of the process C) Entry 10; D) Entry 11; E) Entry 12; F) Entry 13.

HSQC NMR analyses from Lignin^{ox} synthesis study

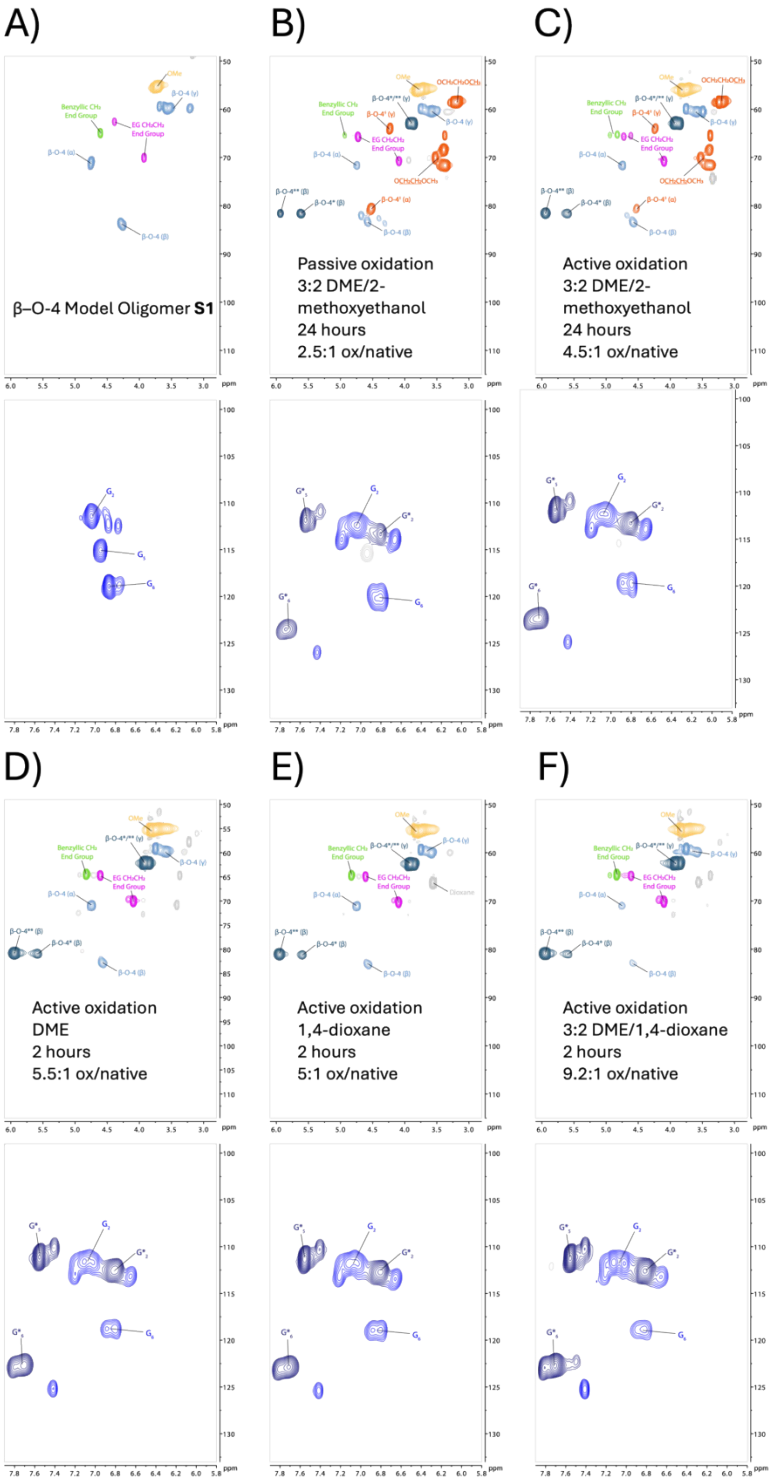


Figure S10. HSQC NMR (700 MHz, DMSO-d_6) analysis of the linkage region (top) and aromatic region (bottom) of A) β-O-4 model oligomer **S1** and B) – F) β-O-4 model oligomer oxidised under various conditions. The ratio of lignin^{ox}:native was determined by the relative integrals for crosspeak signals corresponding to the β-positions. Passive oxidation involved the use of a balloon of oxygen only, whereas active oxidation involved forcing oxygen through the reaction by squeezing multiple oxygen-containing ballons successively into the reaction on a regular basis throughout the reaction.

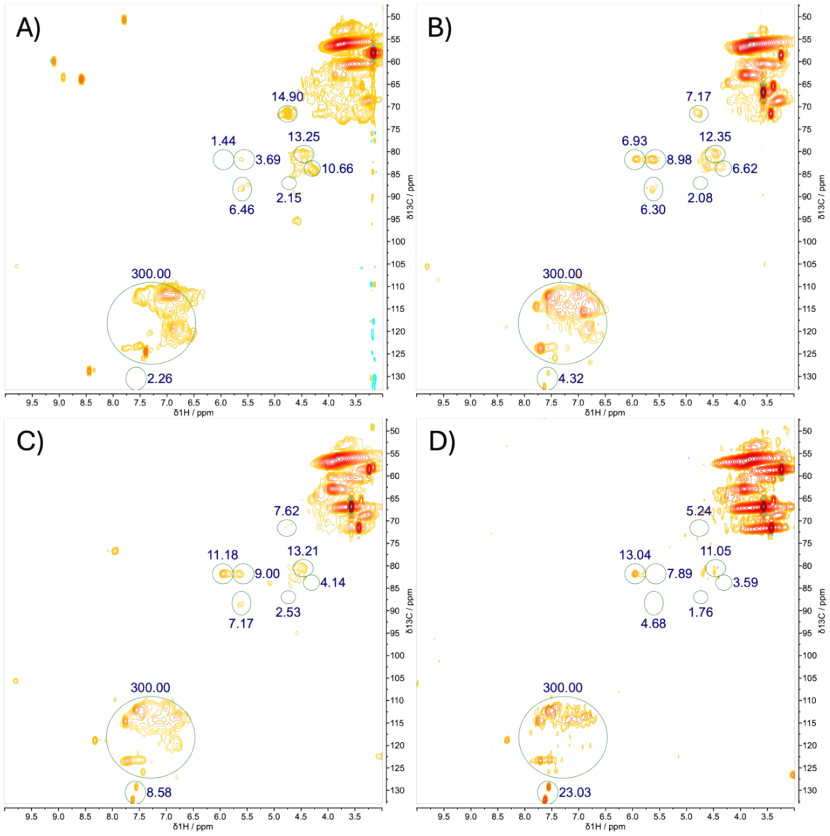


Figure S11. HSQC NMR (700 MHz, DMSO-d_6) analysis of samples from the table in Figure 2f). A) entry 1 catalytic oxidation with DDQ and ^{t}BuONO (15 wt.% of each) at 85 °C in DME/1,4-dioxane (3:2) under 5 atmospheres of compressed air on methylated lignin. B) entry 2 (10% wt. % DDQ and tBuONO) C) entry 3 (25 wt. % DDQ and ^{t}BuONO) D) entry 4 Increasing the concentration that the lignin oxidation reaction was run at from 0.02 g/mL to 0.05 g/mL.

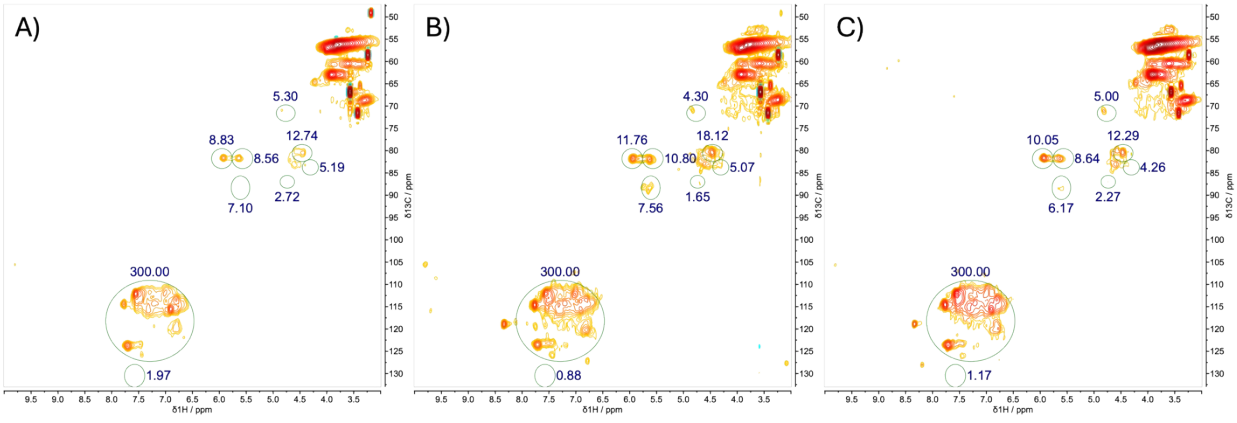


Figure S12. HSQC NMR (700 MHz, DMSO-d_6) analysis of samples from the table in Figure 2f A) entry 5 - further scale up to 2.9 grams of lignin. B) entry 6 - improved by increasing the compressed air pressure to 10 atmospheres. C) entry 7 – a repeat of entry 6 leading to the conclusion that these reaction conditions were reasonably reproducible (entry 7).

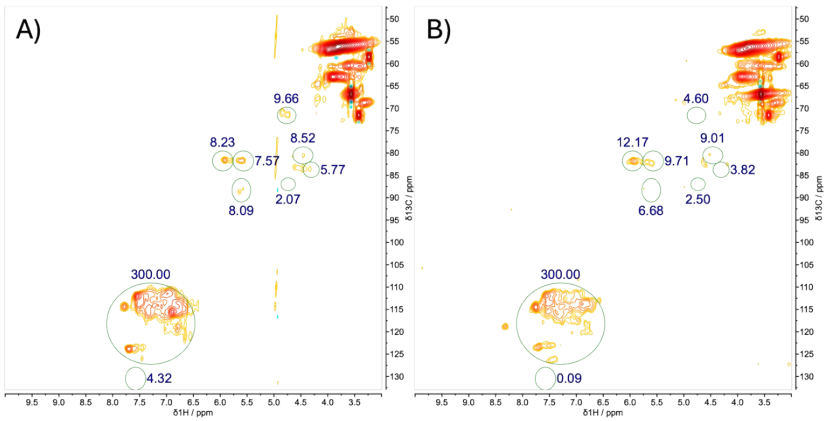


Figure S13. HSQC NMR (700 MHz, DMSO-d_6) analysis of samples from the table in Figure 2f A) entry 8 - in the first of two runs the level of conversion (73%) was slightly lower than hoped for; B) entry 9 - however, an increase in reaction time from 16 to 20 hours (c.f. Figure 2f) entries 8 and 9) delivered a high quality sample of sitka spruce lignin^{OX} on a significant scale in 91 wt. % yield (Figure 2e).

An Alternative Simplified LVF Assessment

During review, an excellent suggestion was made by the Reviewer to simplify further the calculation carried out. The results of this simplification are presented here.

Equation S1

$$\frac{Cost\ of\ Solvent}{\left(\frac{m_{biomass}}{m_{solvent}}\right) x \left(\frac{m_{lignin}}{m_{biomass}}\right)} = \text{Cost per kilogram of lignin prepared / USD/kg}$$

Where Cost of Solvent = Cost of solvent / USD/kg (converted from Yuan/tonne)

$m_{biomass}$ = mass of biomass used / kg

$m_{solvent}$ = mass of alcohol solvent / kg (converted from volume using densities in Table S3)

m_{lignin} = mass of isolated lignin / kg

Table S6. Calculated cost per kilogram of lignin prepared (USD/kg) for selected organosolv pretreatments carried out on Sitka spruce sawdust based on solvent costs and exchange rates on 28th March 2024 and 28th March 2025 using Equation S1.

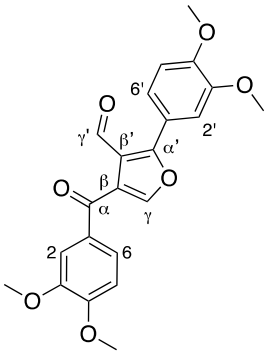
Pretreatment	Cost per kilogram of lignin prepared / USD/kg	
	28th March 2024	28th March 2025
Bronze Methanosolv	86.90	89.28
Bronze Ethanosolv	170.63	145.66
Bronze Isobutanosolv	100.11	96.81
Bronze Butanosolv	84.23	71.48
Bronze Pressurised Methanosolv	28.78	29.57
Bronze Pressurised Ethanosolv	80.35	68.59

The calculated cost per kilogram of the lignins prepared from the different pretreatment protocols follow broadly similar trends to the LVFs calculated (Table 2 in the main text). For example, bronze ethanosolv remains the least competitive with the highest cost per kilogram on both dates. Bronze isobutanosolv affords slightly more expensive lignin than bronze methanosolv, bronze butanosolv and bronze pressurised ethanosolv which are all similar. Bronze pressurised methanosolv gives the cheapest cost of production on both dates but suffers from the same drawback as the original LVF calculation of not considering the significant limitations of a pressurised process compared to an atmospheric pressure process. The same result is observed overall that bronze methanosolv and butanosolv pretreatments give the most economically viable lignins, with only the changing cost of solvent influencing which process should be used. While this methodology simplifies the LVF calculation further, it loses all

molecular detail about the lignin’s structure and so may underperform in predicting the success in a given process.

Identification of product of oxidation of Eudesmin 1 and Yangambin 4

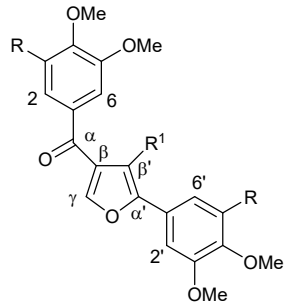
4-(3,4-dimethoxybenzoyl)-2-(3,4-dimethoxyphenyl)furan-3-carbaldehyde (2)



Aldehyde **2** was prepared from Eudesmin **1** (0.15 g, 0.39 mmol, 1.00 eq.) using General Procedure I (Step 1: with 1,4-dioxane (19.5 mL) and DDQ (0.44 g, 1.95 mmol, 5.00 eq.); followed by Step 2 with DCM (19.5 mL) and DMP (0.17 g, 0.39 mmol, 1.00 eq.). Flash column chromatography (0 – 60% EtOAc in petroleum ether) was performed to afford **2** (81.2 mg, 53%) as a yellow amorphous solid.

^{1}H NMR (700 MHz, Chloroform-*d*) δ = 10.26 (s, 1H, Hγ'), 7.77 (d, *J* = 2.1, 1H, H2'), 7.75 (s, 1H, Hγ), 7.68 (dd, *J* = 8.4, 2.1, 1H, H6'), 7.57 (d, *J* = 2.0, 1H, H2), 7.54 (dd, *J* = 8.3, 2.0, 1H, H6), 6.98 (d, *J* = 8.4, 1H, H5'), 6.92 (d, *J* = 8.3, 1H, H5), 4.00 (s, 3H, C4'-OMe), 3.98 (s, 3H, C3-OMe), 3.97 (s, 3H, C4-OMe), 3.96 (s, 3H, C3'-OMe).
^{13}C NMR (176 MHz, $CDCl_3$) δ 188.00 (Cα), 186.29 (Cγ'), 159.94 (Cα'), 153.91 (C3), 151.42 (C3'), 149.47 (C4), 149.01 (C4'), 145.04 (Cγ), 131.23 (C1), 127.02 (Cβ), 124.85 (C6), 121.87 (C6'), 121.24 (C1'), 119.87 (Cβ'), 111.18 (C2, C2'), 111.05 (C5'), 110.08 (C5), 56.31 (C4-OMe), 56.24 (C3-OMe), 56.22 (C3'-OMe), 56.16 (C4'-OMe).
IR (FTIR)ν_{max}: 2970 (br), 2900 (br), 1681 (s), 1635 (s), 1593, 1417 cm^{-1}.
HRMS (NSI+) calculated for $C_{22}H_{21}O_7^+$ $[M+H]^+$ 397.1282; found 397.1282.

(4-(hydroxymethyl)-5-(3,4,5-trimethoxyphenyl)furan-3-yl)(3,4,5-trimethoxyphenyl)-methanone (6) and 4-(3,4,5-trimethoxybenzoyl)-2-(3,4,5-trimethoxyphenyl)furan-3-carb-aldehyde (5)



5 R = OMe, R^1 = CHO
6 R = OMe, R^1 = CH_2OH

To synthesise only aldehyde 5: **5** was prepared from Yangambin **4** (502 mg, 1.12 mmol, 1.00 eqv.) using General Procedure I step 1: With 1,4-dioxane (56 mL) and DDQ (1.02 g, 4.50 mmol, 5.00 eq.) followed by step 2: With DCM (56 mL) and DMP (476 mg, 1.12 mmol, 1.00 eq.). Flash column chromatography (0 – 50% EtOAc in petroleum ether) was performed to afford **5** (199 mg, 39%) as an orange powder. Recrystallisation from EtOAc gave crystals suitable for X-ray crystallographic analysis.

Alcohol 6

Appearance: Yellow amorphous solid, **R.f**. ~0.4 (50% EtOAc in petroleum ether)

^{1}H NMR (500 MHz, Chloroform-*d*) δ = 7.87 (s, 1H, Hγ), 7.18 (s, 2H, H2, H6), 6.90 (s, 2H, H2', H6'), 4.77 (d, *J* = 7.1, 2H, Hγ'), 4.11 (t, *J* = 7.1, 1H, Hγ'-OH), 3.96 (s, 3H, C4-OMe), 3.94 (s, 6H), 3.93 (s, 6H), 3.91 (s, 3H, C4'-OMe).

^{13}C NMR (126 MHz, $CDCl_3$) δ 190.75 (Cα), 153.71 (2C, C-OMe), 153.53, (Cα'), 153.35 (2C, C-OMe), 148.20 (Cγ), 142.78 (C4), 138.97 (C4'), 133.67 (C1), 127.65 (Cβ), 124.89 (C1'), 120.51 (Cβ'), 106.89 (C2, C6), 104.75 (C2', C6'), 61.20 (C4-OMe), 61.15 (C4'-OMe), 56.53 (2C, OMe), 56.40 (2C, OMe), 55.65 (Cγ').

IR (FTIR)ν_{max}: 3662 (br), 2985 (br), 2900, 1627 (s), 1573, 1504, 1411, 1232 (br) cm^{-1}.

HRMS (NSI+) calculated for $C_{24}H_{27}O_9^+$ $[M+H]^+$ 459.1650; found 459.1644.

Aldehyde 5

Appearance: Orange amorphous solid; **R.f**. ~0.3 (50% EtOAc in petroleum ether)

^{1}H NMR (500 MHz, Chloroform-*d*) δ = 10.31 (s, 1H, Hγ'), 7.80 (s, 1H, Hγ), 7.47 (s, 2H, H2', H6'), 7.21 (s, 2H, H2, H6), 3.97 (s, 6H, 2 x OMe), 3.96 (s, 3H, C4-OMe), 3.94 (s, 3H, C4'-OMe), 3.92 (s, 6H, 2 x OMe).

^{1}H NMR (700 MHz, DMSO-d_6) δ = 10.15 (s, 1H, Hγ'), 8.51 (s, 1H, Hγ), 7.39 (s, 2H, H2', H6'), 7.22 (s, 2H, H2, H6), 3.87 (s, 6H , 2 x OMe), 3.86 (s, 6H, 2 x OMe), 3.78 (s, 3H, OMe), 3.75 (s, 3H, OMe).

^{13}C NMR (126 MHz, $CDCl_3$) δ 188.16 (Cα), 186.36 (Cγ'), 159.39 (Cα'), 153.38 (2C, C-OMe), 153.34 (2C, C-OMe), 145.74 (Cγ), 143.12 (C4), 140.62 (C4'), 133.27 (C1), 127.22 (Cβ),

123.56 (C1'), 120.32 (Cβ'), 107.14 (C2, C6), 105.68 (C2', C6'), 61.19 (C4/C4'-OMe), 61.16 (C4/C4'-OMe), 56.56 (2C, *m*-OMe), 56.47 (2C, *m*-OMe).

^{13}C NMR (176 MHz, DMSO) δ 187.45 (Cα), 186.57 (Cγ'), 158.26 (Cα'), 153.02 (2C, Ar-C), 152.84 (2C, Ar-C), 147.36 (Cγ), 142.14 (C4), 139.64 (C4'), 132.48 (C1), 125.80 (Cβ), 123.24 (C1'), 120.31 (Cβ'), 106.84 (C2, C6), 105.46 (C2', C6'), 60.23 (2C, C4, C4'-OMe), 56.11 (2C, *m*-OMe), 56.09 (2C, *m*-OMe).

IR (FTIR)ν_{max}: 2987 (br), 2900, 1799 (s), 1734 (s), 1681 (s), 1645, 1581, 1454, 1232 cm^{-1}.

HRMS calculated for $C_{24}H_{25}O_9^+$ $[M+H]^+$ 457.1493; found 457.1492.

X-Ray Crystallographic Analysis of Compound 5

X-ray diffraction data for compound **5** were collected at 173 K using a Rigaku FR-X Ultrahigh Brilliance Microfocus RA generator/confocal optics [Mo Kα radiation (λ = 0.71073 Å)] with XtaLAB P200 diffractometer. Intensity data were collected using ω steps accumulating area detector images spanning at least a hemisphere of reciprocal space. Data were collected and processed (including correction for Lorentz, polarization and absorption) using CrystalClear.[S7] The structure was solved by direct methods (SIR2011[S8]) and refined by full-matrix least-squares against F^2 (SHELXL-2019/3[S9]). Non-hydrogen atoms were refined anisotropically, and hydrogen atoms were refined using a riding model. All calculations were performed using the Olex2[S10] interface.

Selected crystallographic data for **5**: $C_{24}H_{24}O_9$, M = 456.43, triclinic, a = 9.885(2), b = 10.504(3), c = 10.763(3) Å, α = 78.835(11), β = 87.376(12), γ = 88.318(13) °, Vol. = 1095.0(5) Å^3, T = 173 K, space group $P\bar{1}$ (no. 2), Z = 2, 13488 reflections measured, 3972 unique (R_{int} = 0.0525), which were used in all calculations. The final R_1 [$I > 2\sigma(I)$] was 0.0522 and wR_2 (all data) was 0.1438.

CCDC 2445448 contains the supplementary crystallographic data for this paper. These data can be obtained free of charge from The Cambridge Crystallographic Data Centre via www.ccdc.cam.ac.uk/structures.

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