COMMUNICATION

Enhancing lignin depolymerization via a dithionite-assisted organosolv fractionation of birch sawdust

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Sodium dithionite is utilized as a reducing agent in the organosolv fractionation of lignocellulose to concommitantly produce cellulosic pulp and promote the reductive conversion of lignin into phenolic monomers. Reactions with model compounds highlight the role of sodium dithionite with respect to the reductive cleavage of β-O-4 bonds in lignin and the consequent formation of phenolic monomers.

Introduction

In view of the pressing need for sustainable alternatives to fossil feedstock, lignocellulosic biomass gained considerable attention as a potential source of renewable energy and materials.[1] Recently, within the concept of integrated biorefineries, promising one-step processes were proposed for the simultaneous fractionation of lignocellulose and conversion of lignin toward a handful of phenolics.[2–7] These methods – which target the formation of a lignin oil, rich in phenolic monomers – are usually referred to as "lignin-first biorefineries", to highlight that the valorization of lignin to chemicals is realized upfront. [6–9]

A popular strategy adopted within lignin-first processes is Reductive Catalytic Fractionation (RCF),[10] also referred to as Early-stage Catalytic Conversion (ECCL)[3] or Catalytic Upstream Biorefining (CUB),[11] consisting of an organosolv treatment of lignocellulosic biomass in the presence of a heterogeneous redox catalyst and of a source of hydrogen.[6,10] Under such conditions, labile β-O-4 bonds, the most abundant in lignin structures, are cleaved and the generated lignin fragments are stabilized by catalytic reduction reactions, which prevent re-condensation towards the recalcitrant species usually found in technical lignins. Several authors studied RCF of hardwoods and reported the achievement of outstanding delignification yields (up to ~90%) and phenolic monomers yields (up to 50-60%), as well as high yields of carbohydrate pulps with nearly complete cellulose preservation, amenable toward enzymatic or chemical conversion.[11–15]

Despite providing a powerful means for full valorization of lignocellulose, RCF processes suffer few limitations that complicate their scalability and negatively impact their environmental performance. Notably, the frequent use of costly precious metals and the adoption of high pressures of hydrogen gas constitute primary issues that need to be addressed in order to minimize processing costs as well as safety and equipment requirements.[7,16]

Van den Bosch *et al.* conducted an in-depth study of the role of the solvent, the catalyst and the hydrogen source in the frame of RCF, and pointed out that lignin depolymerization occurs primarily via solvolysis.[17] In particular, the authors postulated the necessity of a reductive step for the solvolytic cleavage of β-O-4 bonds in lignin structures, possibly involving the solvent as a source of hydrogen. Metal-based catalysis is only involved in the final stabilization of the generated reactive lignin moieties, via the hydrogenation of alkylic side chains in lignin units. More recently, Klinger *et al.* demonstrated that reductive cleavage of β-O-4 bonds in pre-extracted, oxidized, lignin can be accomplished using nucleophilic thiols as reducing agents, with the concomitant formation of the correspondent disulfides.[18,19] The authors reported a considerable formation of phenolic monomers and short oligomers as well as a minor incorporation of sulfur in the lignin matrix.

These results prompted us to investigate a modified organosolv process, which could lead to the simultaneous production of cellulosic pulp and conversion of lignin into phenolic monomers, but in the absence of hydrogen gas, heterogeneous catalysts and nucleophilic thiols. Sodium dithionite ($Na_2S_2O_4$), an inexpensive and readily available sulfur-based compound, was selected as a reducing agent. $Na_2S_2O_4$ is already adopted in the pulping industry for the bleaching of pulps[20,21] and was also recently employed in a patented process for the depolymerization of Kraft lignin.[22] However, to our knowledge, there is no record of the adoption of sodium dithionite in the frame of organosolv fractionation of lignocellulose. Yet, the use of $Na_2S_2O_4$ for the depolymerization of lignin directly from native biomass may constitute an interesting route for the valorization of lignin during pulping.

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Herein, we disclose the potential of the dithionite-assisted organosolv fractionation (DAOF) process for the treatment of lignocellulose and we rationalize the role of the reducing agent in the formation of phenolic monomers via reactions with model compounds. We argue that this strategy represents a practical way to modify a pulping process while targeting the reductive depolymerization of lignin for a sustainable production of monoaromatics.

Results and discussion

Dithionite-assisted organosolv fractionation: proof of concept

In order to assess the potential of DAOF, batch experiments were carried out in triplicate treating 3 g of birch sawdust (*Betula pendula,* particle size ≤ 2 mm,[23] biomass composition reported in Table S1) in 120 mL of an equivolumetric mixture of n-butanol and water, in the presence of 1 g of sodium dithionite, at a temperature of 200 °C for a duration of 3 hours. After the fractionation, a solid pulp and two liquid fractions (an organic and an aqueous phase) were collected. An outline of the process is presented in Figure 1. As solvent, the n-butanol – water mixture facilitates the subsequent separation of the isolated fractions.[24–26] Typically, the solid fraction contains the retained polysaccharides (C5, C6), the organic liquid fraction contains lignin-derived phenolics, and the aqueous liquid fraction comprises solubilized non-condensed C5 and C6 derivatives (e.g. mono- and oligosaccharides, polyols and organic acids). The obtained product streams were then extensively characterized by a series of analytical procedures (Figure S1).

The solid fraction

The light brown solid isolated after the fractionation was analyzed for dry matter (DM) and ash (Figure S2),[27,28] then subjected to acid hydrolysis,[29] which revealed that about 91.7% of C6 polysaccharides initially contained in the biomass were recovered in the solid fraction (Table 1). In addition, 81.9% of C5 polysaccharides and 71.6% of the acid-insoluble lignin (AIL) contents were removed, leading to a cellulosic pulp with a high cellulose purity of 76.0%. The obtained pulp was analyzed via X-ray diffraction (Figure S3), and cellulose crystallinity index (CI) was determined by Segal's method.[30] Despite the extensive

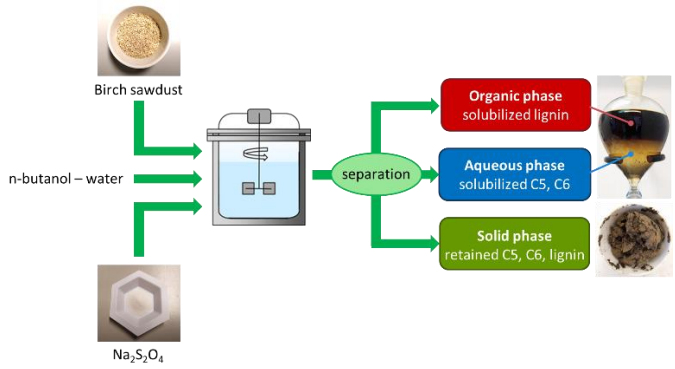


Figure 1 Outline of the dithionite-assisted organosolv fractionation of birch sawdust.

Table 1 Characterization of the solid and liquid fractions obtained for the treatment of birch sawdust (3 g, particle size < 2 mm) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N2 (introduced at ambient temperature), in the presence of $Na_2S_2O_4$ (1 g), for a duration of 3 hours.

Solid fraction	Recovery (wt%)	C5	18.1 ± 3.2
		C6	91.7 ± 1.3
		Lignin[a]	28.4 ± 2.7
	Conversion (%)	Glucan	91.4 ± 4.6
		Xylan	92.8 ± 4.0
	CI		54.4 ± 3.5
Organic fraction	Yield (wt%)	Lignin oil[b]	93.4 ± 6.8
		Monophenolics[b]	18.1 ± 1.7
		C5, C6 derivatives[c]	0.1 ± 0.0
Aqueous fraction	Yield (wt%)	Lignin oil[b]	3.2 ± 0.4
		Monophenolics[b]	0.5 ± 0.1
		C5, C6 derivatives[c]	7.5 ± 0.4

[a] Acid-insoluble lignin

[b] Expressed with respect to the weight of acid-insoluble lignin contained in the initial biomass.

[c] Non-condensed carbohydrate derivatives, expressed with respect to the total weight of polysaccharides contained in the initial biomass.

removal of hemicellulose and lignin achieved during the fractionation, which was reported to favor increased CI,[31,32] no major difference was observed between the CIs of the untreated birch sawdust and that of the pulp, 48.7% and 54.4%, respectively (Table 1). Partial cellulose depolymerization[27] or re-deposition of pseudolignin[28] (also termed "humins") on cellulose fibers may constitute possible explanations for the fact that the measured CI increase remained so minor.

The morphological features of pristine biomass and of the obtained pulp were further inspected via scanning electron microscopy (SEM). While the surface of birch sawdust appeared rough and compact (Figure S4a), after treatment, the pulp possessed a considerably smoother surface and bundles of fibers with a diameter of about 10 µm could be observed (Figure S4b). At higher magnification, thin fibers possessing diameters of about 1 µm were apparent (Figure S4c), pointing at the partial disassembly of fibrous bundles achieved during the treatment, which is expected to favor further enzymatic conversion of the pulp.[33]

In order to get a better insight into the susceptibility of the cellulosic pulp toward further processing, it was subjected to enzymatic digestion (Table 1). A substantial enhancement of both glucan and xylan convertibility to their corresponding monosaccharides was observed for the solid pulp (91.4% and 92.8%, respectively) compared to untreated biomass (8.3% and 9.7%, respectively), confirming that the structural changes occurring in the lignocellulose matrix during fractionation ultimately favor the pulp processability. Interestingly, analogous *blank* organosolv experiments carried out in the absence of sodium dithionite produced a solid pulp with very similar properties (Table S2), indicating that the use of dithionite does not affect the pulp composition, CI and enzymatic digestibility. On the other hand, a relevant difference

between DAOF and a *blank* organosolv treatment was found in the degree of polymerization (DP) of cellulose in the isolated pulps, which was estimated via viscosimetric analysis, according to the NF G 06-037 norm (Table S3). Notably, DAOF produced a pulp with a DP_v of about 780 AGU, considerably larger than that of the *blank* organosolv pulp (~230 AGU). We reason this difference may be due to the higher acidity of the medium in the *blank* organosolv experiment (see below), which may favor cellulose depolymerization.

The organic fraction

The organic liquid fraction obtained from the process was analyzed for dry matter and ash (Figure S2),[27,28] then a lignin oil was isolated from the organic fraction by drying a sample and subjecting it to liquid – liquid extraction with dichloromethane and water, following a procedure reported elsewhere.[12,17] Drying the dichloromethane extract yielded a viscous brown lignin oil, corresponding to about 93.4% of the weight of AIL contained in the initial biomass (Table 1). Interestingly, this value is higher than the 71.6% measured for lignin removal in the pulp, suggesting the possible redeposition of pseudolignin on the pulp or the incorporation of (hemi)cellulose derivatives in lignin oil. Indeed, humic products, often generated during hydrothermal processing of lignocellulosic material via condensation of dehydration products from polysaccharides,[34] are known to be partially soluble in alcohols and dichloromethane.[35] Thus, we reason that humins are likely responsible for the apparent generation of lignin during the fractionation.

The molecular weight distribution (MWD) of the lignin oil was evaluated via gel permeation chromatography (GPC), and compared to that obtained for a *blank* organosolv reaction, carried out in the absence of sodium dithionite (Figure 2). A broad but small peak was detected at a molecular weight below 100 g mol^{-1} for the DAOF experiment, which may be related to degradation products from the solvent and the reducing agent.

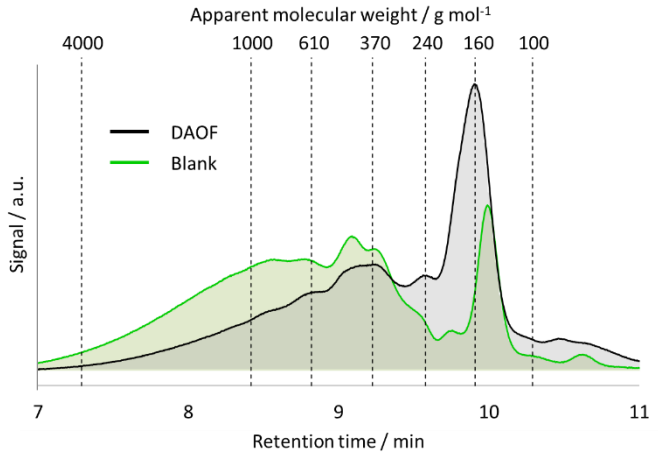


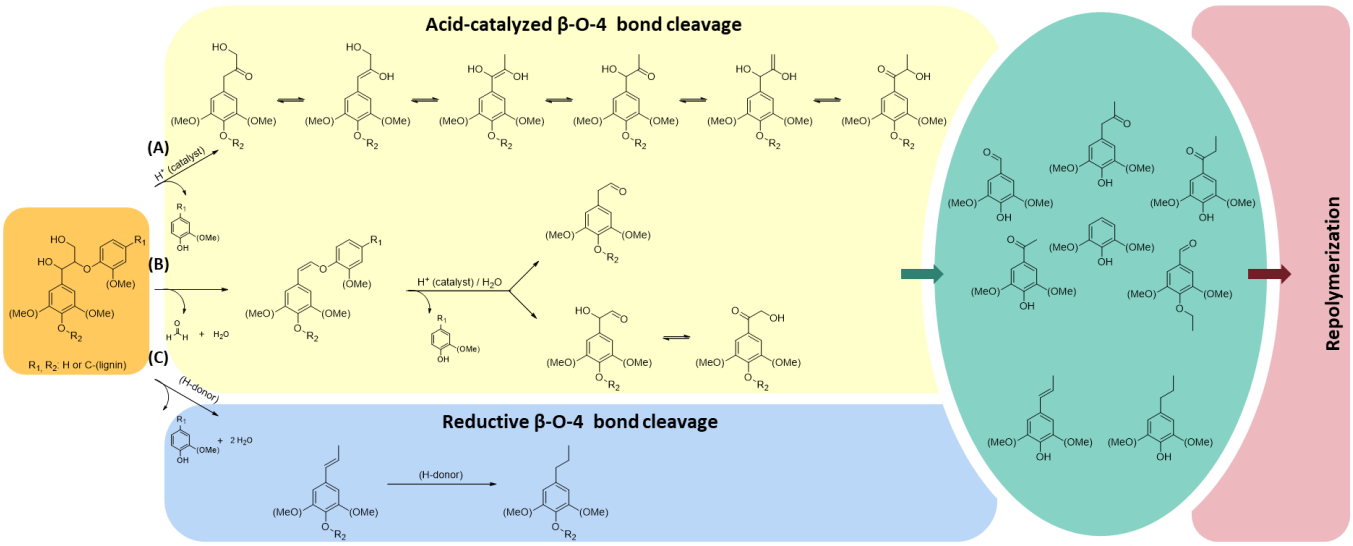
Figure 2 Comparison of the GPC chromatograms of the lignin oil isolated from the organic fraction obtained after treating birch sawdust (3 g, particle size < 2 mm) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature), for a duration of 3 hours, in the absence (blank) and in the presence of $Na_2S_2O_4$ (1 g). The total surface of each chromatogram was normalized with respect to the yield of lignin oil obtained for the corresponding experiment.

Two major peaks were found at about 160 g mol^{-1} and 240 g mol^{-1}, corresponding to phenolic monomers. These constitute the major fraction of the chromatogram, indicating the formation of monophenolics during the fractionation process. In addition, two smaller peaks were observed at 370 g mol^{-1} and 610 g mol^{-1}, likely corresponding to phenolic dimers and trimers. These were followed by a tail extending to higher molecular weights, which reveals the presence of some condensation products (lignin oligomers and humins). For the *blank* experiment, the peak corresponding to phenolic monomers was much smaller, and, instead, the presence of peaks at higher molecular weights was substantially larger than in the DAOF process. Also, the tail extending to even higher molecular weight products was more pronounced in the case of the *blank* experiment. Thus, overall, the analysis of the MWD of lignin oil unambiguously demonstrates that the introduction of sodium dithionite within the organosolv process leads to enhanced depolymerization of the isolated lignin, with an increased production of phenolic monomers.

The monomers composition of the lignin oil was analyzed by gas chromatography (GC-MS and GC-FID). The chromatogram reported in Figure S5a and the data summarized in Table S4 show that several monophenolic compounds are present in the lignin oil, accounting for a total monomer yield of 18.1% (based on the content of AIL in the initial biomass). The most abundant phenolic monomers that were detected are: 4-ethoxy-3,5-dimethoxybenzaldehyde (4.3%), 4-propenyl syringol (4.3%), desaspidinol (3.2%) and acetosyringone (2.7%). The formation of phenolic compounds possessing aldehyde or ketonic groups within the alkylic side chains is typical of acid-catalyzed organosolv depolymerization of lignin,[3] while the formation of monomers with unsaturated alkylic side chains was postulated or demonstrated to occur under RCF conditions.[14,17,36] Therefore, we reason that lignin depolymerization during DAOF may follow a combination of acid-catalyzed and reductive pathways for the cleavage of β-O-4 bonds in lignin and for the generation of phenolic monomers (**Erreur ! Source du renvoi introuvable.**). However, the formation of structures with an alkylic side chain containing four carbon atoms, such as desaspidinol, is somewhat surprising (see below).

For comparison, the analysis of phenolic monomers in the lignin oil isolated from *blank* organosolv reactions performed in the absence of dithionite revealed a considerably lower formation of monophenolics (total monomer yield of 3.6%, see Table S5), corroborating the deductions made based on GPC. Notably, marginal formation of sinapyl alcohol occurred for the *blank* organosolv experiments, while such monophenolic with unsaturated alkyl side chain possessing a terminal hydroxyl group was not observed in DAOF.

To inspect the presence of organic acids and dehydration products from carbohydrates, a sample of the organic phase was subjected to HPLC analysis. The chromatogram in Figure S6a and the data in Table S6 show that 0.1% of formic acid and 0.9% of acetic acid were detected (on a DM basis), indicating a minimal presence of non-condensed carbohydrate derivatives in the organic phase. A similar outcome was observed also for the *blank* organosolv experiment.



Scheme 1 Possible pathways leading to the formation of phenolic monomers during organosolv fractionation in the presence of sodium dithionite. Pathways (A) and (B) rely on the acid-catalyzed cleavage of β-O-4 bonds,[3,4] while the pathway (C) consists of a reductive cleavage of ether bonds.[17] The combination of acid-catalyzed and reductive lignin depolymerization pathways may explain the different structures observed for the phenolic monomers produced during the process (enclosed in the green oval). The generated monomers can subsequently undergo repolymerization at an extent that depends on reaction conditions.

The aqueous fraction

The aqueous liquid fraction obtained after DAOF was analyzed for dry matter and ash (Figure S2),[27,28] then a lignin oil was isolated, corresponding to about 3.2% of the weight of AIL contained in the initial biomass (Table 1). GC analyses (Figure S5b and Table S4) showed that the total yield of monophenols was as low as 0.5%. These findings demonstrate that efficient isolation of lignin derivatives in the organic phase was achieved. In order to inspect the presence of non-condensed derivatives that may arise from the decomposition of C5 and C6 polysaccharides (mono- and oligosaccharides, polyols and organic acids), a sample of the aqueous phase was subjected to HPLC analysis. Xylose, 1,2-propanediol and formic acid are the major carbohydrate derivatives that were identified, respectively, accounting for 0.1%, 0.7% and 3.7% of the initial biomass DM (Figure S6b and Table S6). These components were reported to arise from the decomposition of (hemi)cellulose during hydrothermal treatment, via hydrolysis, retro-aldol and dehydration pathways.[37–39] The presence of a large amount of formic acid in the aqueous phase is remarkable. In addition, acetic acid (6.5%) was detected in the aqueous phase, likely generated from the deacetylation of hemicellulose.[40,41] The formation of organic acids during the process is known to contribute to lowering the pH of the medium and to enhancing acid-catalyzed cleavage of ether and ester bonds in the lignocellulose structure.[42]

Blank organosolv experiments, carried out in the absence of dithionite, led to the formation of lower amounts of formic and acetic acid (0.5% and 3.1%, respectively), and a larger yield of xylose (1.2%), indicating that the presence of $Na_2S_2O_4$ affects (hemi)cellulose decomposition. Despite the larger production of formic and acetic acid observed during DAOF, the pH value of the aqueous phase was larger in this case, compared to that measured after a *blank* organosolv treatment (about 5.9 and 3.3, respectively), highlighting how the presence of dithionite leads to a lower acidity of the medium.

The origin of desaspidinol

The generation of phenolic compounds with alkylic side chains containing four carbon atoms within DAOF is rather unexpected since these structures are not present in native lignin.[3] On the other hand, desaspidinol was reported to form in the frame of the catalytic hydrogenolysis of lignin, and a correlation between its production and the use of formic acid as a solvent for lignin depolymerization was recognized.[43–47] Therefore, we surmise that, in DAOF, carbohydrate derivatives, such as formic acid, produced by the decomposition of (hemi)cellulose may lead to the subsequent formation of desaspidinol.

To shed light on the role of carbohydrate derivatives with respect to the formation of desaspidinol during the fractionation, milled wood lignin (MWL) was extracted from birch sawdust and subjected to an analogous process. GC analysis of the isolated lignin oil (Table S7), showed that the total yield of phenolic monomers was more than two times lower than that observed for DAOF of lignocellulosic biomass, possibly due to partial modifications of the native lignin structure during MWL extraction. A significant finding is that the yield of desaspidinol was about 25 times lower for the MWL, which unambiguously confirms that the formation of this monophenol is related to the presence of carbohydrate derivatives in the reaction mixture. A similar experiment was performed treating MWL in the presence of formic acid. In this case, the yield of desaspidinol increased slightly, even if it remained much lower than in DAOF of lignocellulosic biomass. Thus, while formic acid appears to play a role in the formation of desaspidinol during DAOF, other (hemi)cellulose derivatives produced during the fractionation may also influence this phenomenon.

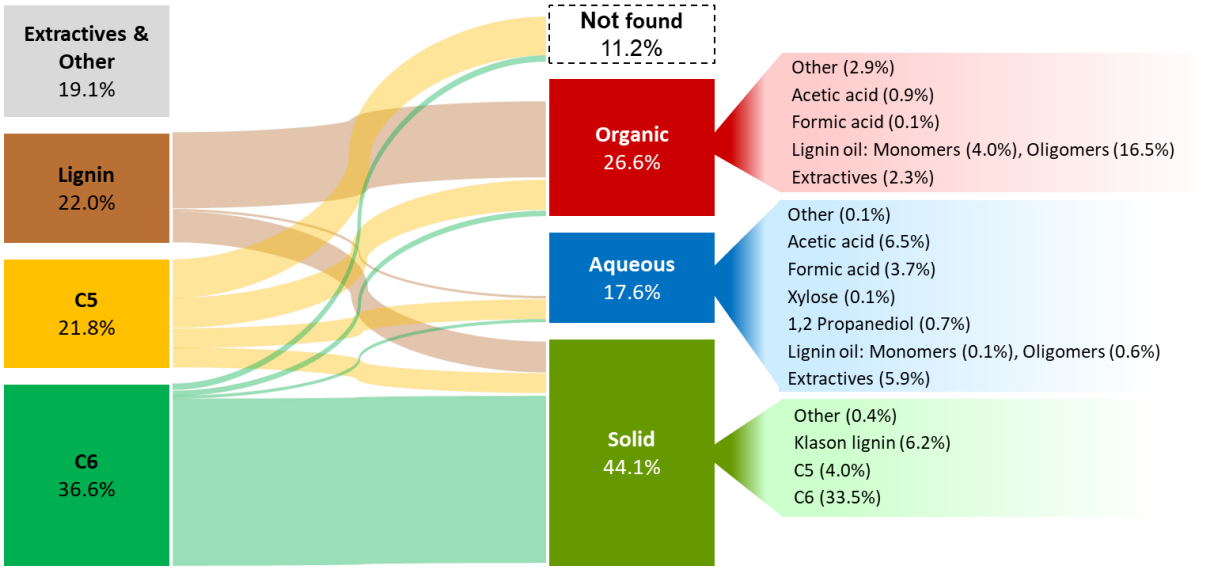


Figure 3 Schematic view of the mass balance (based on DM of the biomass introduced) for biomass components, obtained for the treatment of birch sawdust (3 g, particle size < 2 mm) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature), in the presence of $Na_2S_2O_4$ (1 g), for a duration of 3 hours. The reaction was carried out in triplicates. The ash fraction was not taken into consideration in the mass balance, since it only accounts for a marginal fraction of DM in biomass (0.5%), while most of the ash content in the reaction products derives from $Na_2S_2O_4$. Lignin oil, besides phenolic monomers, was assumed to be composed only of oligomers.

Overview of the fate of biomass components, dithionite and butanol

The compositional investigation of the solid, the organic and the aqueous fractions obtained from DAOF gave insight into the fate of biomass components during processing. An overall characterization of biomass derivatives in the product streams is presented in Table S6, and, based on that, an overall mass balance for lignocellulose components is illustrated in Figure 3. About 89% of the DM initially introduced in the reactor was recovered in the output streams; losses are presumably due to the partial conversion of biomass to volatile components. On the one hand, a relevant amount of C5 derivatives was found in all the isolated fractions, also substantially contributing to material losses. On the other hand, C6 polysaccharides and lignin appeared to be mostly recovered in the solid and organic fraction, respectively, showing that efficient separation of cellulose and aromatics was achieved, which conveniently facilitates subsequent downstream operations.

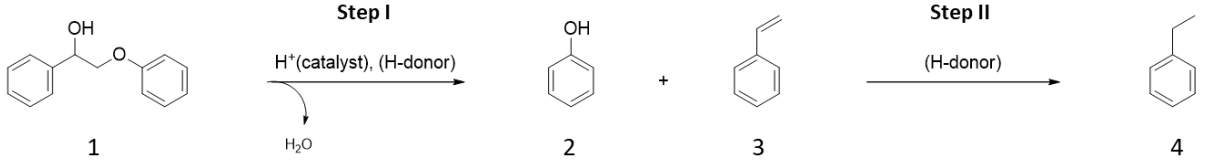
Incorporation of dithionite derivatives in the output streams obtained from the fractionation was assessed by quantification of the sulfur content in each fraction via ICP-AES (Table S8). About 50.5% of the sulfur introduced in the process was recovered in the organic fraction, which points out the formation of organosulfur compounds during the process, such as (poly)sulfides, sulfonates or thiols. An additional 27.4% of sulfur was found in the aqueous fraction, corresponding to inorganic derivatives of dithionite, such as sulfate, sulfite and thiosulfate. The solid fraction accounted for 4.0% of the sulfur introduced, possibly owing to the production of elementary sulfur or to the incorporation of sulfur in the re-deposited pseudolignin. Notably, nearly 18% of the sulfur introduced was not recovered in the product streams, suggesting the formation of volatile sulfur species during DAOF.

The stability of n-butanol during the process was assessed by analyzing its recovery in the liquid fractions via HPLC, which revealed that about 97% of the butanol co-solvent was recovered in the output streams (Table S9), with 93% of it being recovered in the organic phase, indicating that no relevant decomposition of n-butanol occurs during the process.

The role of sodium dithionite explained with model compounds

In order to gain a better insight into the role of dithionite within lignin depolymerization, 2-phenoxy-1-phenylethanol (**1**) was selected as a model compound for β-O-4 bonds in lignin,[48,49] and subjected to similar process conditions. Scheme 2 illustrates the two-step reaction, involving an initial cleavage of **1** (Step I) to generate styrene (**2**) and phenol (**3**), followed by the hydrogenation of styrene to form ethylbenzene (**4**) (Step II).

In the absence of dithionite, the conversion of **1** was low and mostly high molecular weight products were formed, as suggested by the long tail after the peak at about 190 g mol^{-1}, corresponding to the dimer (Figure 4a). In contrary, when the reducing agent was introduced, a greater conversion of **1** was



Scheme 2 Reductive cleavage of 2-phenoxy-1-phenylethanol in the presence of $Na_2S_2O_4$ (Step I) and subsequent hydrogenation of styrene to ethylbenzene (Step II).

observed and another peak at about 110 g mol^{-1} appeared, which became larger as the loading of dithionite was increased, confirming that an excess of sodium dithionite substantially boosted the cleavage of β-O-4 bonds and the formation of monomeric products. The tail extending to high molecular weights appeared less pronounced in the presence of dithionite. However, the small peak at 290 g mol^{-1} indicates that repolymerization reactions were not totally suppressed in the presence of dithionite.

The identification and quantification of the monomeric reaction products (Figure 4b) corroborates the deductions made based on GPC. The addition of sodium dithionite boosted conversion and triggered the formation of **2** and **3**. As the loading of reducing agent was increased, both the dimer conversion and the monomers yields increased. The lack of selectivity toward monomers constitute additional indication that a portion of **1** underwent repolymerization. Furthermore, at high loadings of dithionite, **4** was found as well, showing that the reducing agent can trigger the hydrogenation of alkylic side chains in the monomeric species generated via reductive cleavage of β-O-4 bonds.

A similar set of experiments was performed using 4-propenyl guaiacol (**5**) as a model substrate, a phenolic monomer that was actually generated during DAOF of birch sawdust. In this case also, the conversion increased considerably with the introduction of the reducing agent (Figure S7). Similarly, a small amount of 4-propyl guaiacol (**6**) was formed, pointing again to the ability of dithionite to saturate the alkylic side chains in monophenolics.

No phenolic monomers with propanol-side chains were detected in the reaction products from DAOF, even though these are normally found in native lignin structures,[3] and were also observed in marginal amounts in *blank* organosolv experiments. Thus, to inspect whether the reducing agent is responsible for the cleavage of the terminal hydroxyl group in monolignols' alkylic side chains, an analogous set of experiments was carried out using 3-phenyl-1-propenol (**7**) as a model substrate. When no dithionite was added to the reaction mixture, **7** reacted with n-butanol to form 3-butoxy-propenyl benzene (Figure S8). In contrast, in the presence of the reducing agent, the ether was not formed and the conversion of **7** to 3-phenyl-1-propanol (**8**) was observed, highlighting again the reducing effect of dithionite on the alkyl chain. Interestingly, no other monomers were found, implying that the presence of dithionite does not determine *per se* the cleavage of the hydroxyl group in the side chains of phenolic monomers. Otherwise, it would be expected to be depleted by reaction with the alcohol co-solvent (n-butanol); the latter was in fact almost completely recovered in the output liquid streams. We surmise that the loss of terminal hydroxyl groups in monophenolic products could occur through dehydration reactions in the conditions of DAOF. The resulting unsaturated side chains may eventually be further reduced by dithionite.

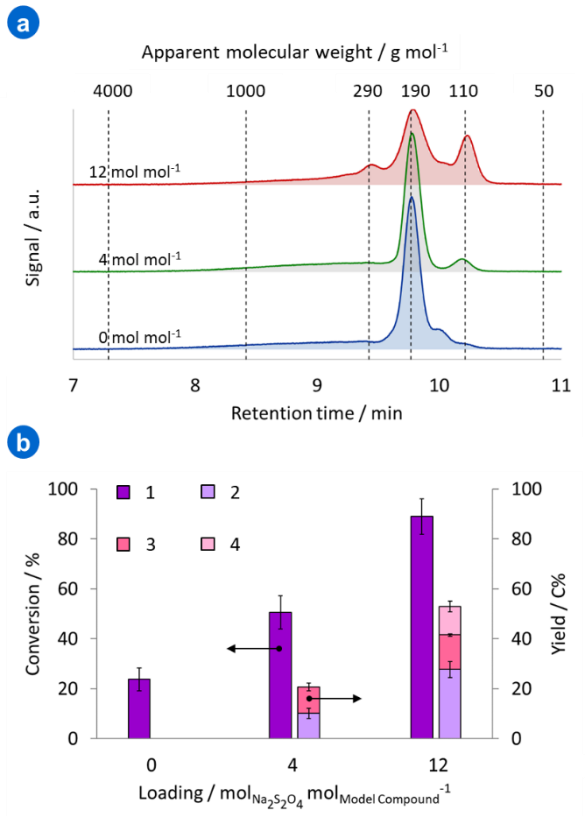


Figure 4 Effect of $Na_2S_2O_4$ on the conversion of 2-phenoxy-1-phenylethanol. (a) Molecular weight distribution of the products determined by GPC and (b) conversion and yield of monomers determined by GC-MS/FID. The reactions were performed in duplicates, treating 0.02 g of 2-phenoxy-1-phenylethanol in a mixture of n-butanol and water (120 mL, 50% v/v), at 250 °C, under 30 bar of N_2 (introduced at ambient temperature), for a duration of 3 hours. The arrows in the figure indicate the reference axis.

Conclusions

A dithionite-assisted organosolv fractionation process is presented, for the concomitant production of cellulosic pulp and the reductive conversion of lignin into phenolic monomers and short oligomers. The process relies on the treatment of lignocellulosic biomass in an equivolumetric mixture of n-butanol and water, in the presence of sodium dithionite as a reducing agent. The immiscibility of n-butanol and water at ambient conditions facilitates product separation. Highly processable cellulose is recovered in the solid, while lignin and humins migrate to the organic phase and non-condensed carbohydrate derivatives are collected in the aqueous phase. Importantly, as compared to classical organosolv conditions, the dithionite-assisted process reaches enhanced lignin depolymerization efficiency and much higher phenolic monomers yields, potentially offering an economically viable alternative to conventional pulping processes (see ESI for a simplified economic assessment). Apart from the monomers classically found in similar processes, the formation of desaspidinol was highlighted and explained by the action of (hemi)cellulose derivatives formed during processing, such as formic acid.

The study of reactions with model compounds revealed that sodium dithionite favors the reductive cleavage of β-O-4 bonds and the hydrogenation of alkylic side chains in the produced

monomers, without, however, suppressing totally the occurrence of repolymerization reactions.

While the operating conditions should be further optimized to exploit the full potential of the method, this first study shows that dithionite-assisted organosolv fractionation (DAOF) is an attractive process for implementing lignin valorization during pulping, in the perspective of developing an integrated biorefinery approach that does not require the use of a catalyst or hydrogen gas to achieve partial depolymerization of lignin.

Experimental section

A complete list of the materials used throughout the work and a detailed description of the experimental procedures are available in the ESI.

Silver birch (*Betula pendula*) wood was collected in Belgium, milled (particle size ≤ 2 mm) and dried in air at ambient conditions. The biomass composition was assessed according to well-established protocols developed by the National Renewable Energy Laboratory (NREL).[27–29,50] Namely, dry matter and ash contents were determined by drying and ashing biomass. The contents of water- and ethanol-soluble extractives were measured by subjecting biomass to two consecutive extractions with water and ethanol, using a Soxhlet extractor. Quantification of structural carbohydrates and lignin was performed by subjecting the extracted biomass to a two-step acid hydrolysis in sulfuric acid.

DAOF experiments were carried out in a 300 mL Parr batch reactor (Parr Instrument Company, Moline, IL, U.S.): 3 g of biomass were introduced in the reactor, together with 60 mL of n-butanol, 60 mL of water and 1 g of sodium dithionite. Air in the reactor was displaced by purging with N_2, then the reactor was pressurized with 30 bar of N_2. The impeller speed was set to 750 rpm and the temperature was increased at a rate of about 10 °C min^{-1}, up to 200 °C. Once the temperature setpoint was reached, the mixture was left to react for a duration of 3 hours. Subsequently, the reactor was cooled down to ambient temperature by letting water flow through the cooling coil, depressurized, and its content was collected. Centrifugation of the product mixture led to the recovery of a solid and a liquid fraction. The solid pulp obtained from the process was washed twice with n-butanol (40 mL) and water (40 mL) to remove polar and apolar components weakly adsorbed to the solid matrix, before being dried at 60 °C to a constant weight. Thereafter, the washing solvents were added to the liquid fraction, and the mixture was filtered to eliminate residual solid particles. The filtrate was transferred to a separating funnel, where it separated into an organic and an aqueous liquid phase, which were subsequently collected.

The dry matter and ash contents of all the isolated fractions were determined according to NREL protocols.[27,28] The solid fraction was analyzed for carbohydrates and lignin, following the same two-step acid hydrolysis procedure used for biomass characterization. In addition, the pulp was characterized via X-ray diffraction analysis, to determine cellulose crystallinity (measured according to Segal's method),[30] via scanning electron microscopy analysis, via viscosimetric analysis to determine the degree of polymerization of cellulose (according to the NF G 06-037 norm), and via enzymatic saccharification, performed following NREL procedures.[51]

Portions of the organic and aqueous liquid fractions were dried under nitrogen flow and subjected to a three-fold liquid-liquid extraction with dichloromethane and water, to isolate lignin derivatives from more polar products. The three dichloromethane fractions were mixed, then dried under vacuum to yield a viscous brown lignin oil that was subsequently analyzed via GPC to inspect its MWD. In order to assess the phenolic monomers composition of lignin oil, a weighed amount of 2-isopropylphenol (internal standard) was added to the dichloromethane extract, which was then analyzed via GC-MS and GC-FID.

Furthermore HPLC-RID analysis of the organic and aqueous fractions was performed to quantify their content of mono- and oligosaccharides, polyols and organic acids, as well as to determine the recovery of the butanol co-solvent in the liquid fractions.

The presence of derivatives of sodium dithionite in the output streams obtained after fractionation was assessed by determination of the sulfur content in each fraction via ICP-AES analysis.

Milled wood lignin was prepared according to a well-established procedure.[52] Birch sawdust underwent Soxhlet extraction as described above, then was milled (800 rpm, 12 hours) using an Emax ball mill (Retsch, Haan, Germany) to yield a fine powder. Milled wood was dispersed in aqueous dioxane (96% v/v) for 48 hours, recycling the wood into fresh solvent after 24 hours. The vacuum-dried extract was dissolved in acetic acid (90% v/v), precipitated in water and air dried, before being dissolved again in 1,2-dichloroethane – ethanol (2:1 v/v), precipitated in ethyl ether and dried to yield MWL. The isolated lignin was then subjected to DAOF as previously illustrated for lignocellulosic biomass. For an optimal reproducibility of the reaction conditions between experiments with biomass and MWL, the amount of MWL employed equaled that of AIL contained in the biomass (about 0.63 g). The treatment of MWL in the presence of formic acid was carried out with an amount of formic acid equal to that obtained after DAOF of raw biomass (about 0.11 g).

The reactions with model compounds were carried out using an identical set-up as that adopted for DAOF experiments with biomass. A weighed amount of the model compound (0.02 g for 2-phenoxy-1-phenylethanol, 0.05 g for 4-propenyl guaiacol and 3-phenyl-1-propenol) was introduced in the reactor, together with 60 mL of n-butanol, 60 mL of water and a chosen amount of sodium dithionite (between 0 and 12 molar equivalents). Subsequently, the reaction and the analysis of the products were performed as described above. For the treatment of 2-phenoxy-1-phenylethanol, a higher reaction temperature of 250 °C was adopted, to promote the cleavage of the dimer.[48]

Author Contributions

We strongly encourage authors to include author contributions and recommend using CRediT for standardised contribution

descriptions. Please refer to our general author guidelines for more information about authorship.

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Conflicts of interest

There are no conflicts to declare.

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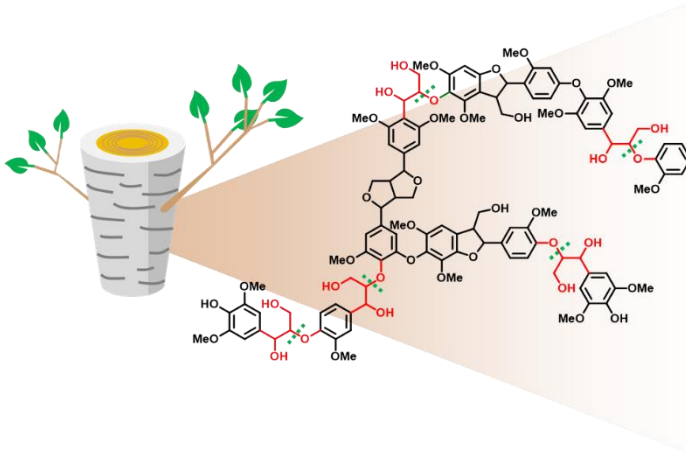

Electronic Supporting Information

Enhancing lignin depolymerization via a dithionite-assisted organosolv fractionation of birch sawdust

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Contents

Chemicals and materials

All commercial chemicals were used without further purification. 2-Isopropyl phenol (98%), 2-methoxy-4-methylphenol (≥98%), 4-methyl-2,6-dimethoxyphenol (≥97%), 2-methoxy-4-propylphenol (≥99%), p-cresol (99%), 4-propylphenol (99%), 3-phenyl-1-propanol (98%), ethylbenzene (99.8%), cinnamyl alcohol (98%), 4-hydroxy-3-methoxyphenylacetone (96%), 4'-hydroxy-3'-methoxyacetophenone (98%), styrene (≥99%), 2-phenoxy-1-phenylethanol, phenol (≥99%), glutaric acid (99%), levulinic acid (98%), malonic acid (99%), methylmalonic acid (99%), Succinic acid (≥99.5%), 1,2-propanediol (≥99.5%), 1,3-propanediol (98%), ethylene glycol (≥99%), furfural (99%), maltose (≥99%), acetic acid (≥99%), DL-lactic acid (85%), crotonic acid (98%), D-gluconic acid (99%), citric acid monohydrate (≥99%), sodium citrate dihydrate (≥99%), sodium dithionite (≥85%) and cellulase enzyme blend were purchased from Sigma Aldrich. Ethanol absolute (100%), tetrahydrofuran (100%), 1,4-dioxan (99.9%), dichloromethane (100%), diethyl ether (100%), 1,2-dichloroethane (≥99.5%), methanol (100%), 2-propanol (99.9%), nitric acid (69%), hydrogen peroxide (30%) and sulfur standard solution (1000 mg/L) were purchased from VWR. (D-)-Mannitol (≥99%), (D+)-glucose monohydrate (≥99%), saccharose (≥99.5%) formic acid (≥98%) and sodium azide (99%) were purchased from Merck Millipore. D-(+)-Xylose (>99%), D-(+)-galactose (99+%), D-(+)-mannose (99+%), DL-malic acid (99%) and oxalic acid (99%) were purchased from Janssen. 4-Ethylphenol (97%), 2-methoxyphenol (98+%), 2,6-dimethoxyphenol (99%), 4-ethyl-2-methoxyphenol (98%), 4-hydroxy-3,5-dimethoxybenzyl alcohol (97%), isoeugenol, cis + trans (98+%), eugenol (99%), 4'-hydroxy-3',5'-dimethoxyacetophenone (97%), 4-allyl-2,6-dimethoxyphenol (98%), 3,4,5-trimethoxytoluene (98%), syringaldehyde (98+%), 5-hydroxymethyl-2-furaldehyde (97%), xylitol (99%), D-(-)-arabinose (99%), 1-butanol (99%) and butyric acid (99+%) were purchased from Alfa Aesar. 4-(3-Hydroxypropyl)-2-methoxyphenol (≥98%) was purchased from TCI. Silver birch (*Betula pendula*) wood was collected in Belgium in 2018.

Biomass preparation

The wood was chopped, milled and sieved to obtain chips with a maximum size of 2 mm, suitable for the fractionation process, which were left to dry in air, then were stored in plastic buckets at ambient conditions.

The milled, air-dried biomass was then analyzed following standard protocols for the characterization of lignocellulosic material, including determination of dry matter and ash content, determination of extractives (water- and ethanol-soluble fractions), structural carbohydrates and acid insoluble lignin.

Analysis of dry matter and ash

Dry matter (DM) and ash of the raw biomass were determined according to National Renewable Energy Laboratory (NREL) protocols (NREL/TP-510-42621 and NREL/TP-510-42622).[1,2] Therefore, a sample of lignocellulosic material (0.5 – 2 g) was weighed in a ceramic crucible and dried at 105 °C to a constant weight to measure the dry matter content. The total ash content was then determined by ashing the dried biomass in a furnace at 575 °C.

Analysis of extractives

Determination of extractives was carried out according to established NREL procedures (NREL/TP-510-42619).[3] Therefore, biomass was milled to obtain a powder (particle size ≤ 0.5 mm). A sample (~5 g) was introduced into an alundum thimble and subjected to two consecutive extractions, first with pure water, then with pure ethanol, using a Soxhlet extractor. Each extraction was carried out for 10 hours, with 4-5 siphon cycles per hour for water and 5-6 siphon cycles per hour for ethanol (siphon cycle = total reflux of the solvent). After the extraction, the water and ethanol extracts were collected separately and dried to a constant weight at 60 °C, then for 1 hour at 105 °C, to determine the amount of water and ethanol extractives.

Analysis of structural carbohydrates and acid insoluble lignin

Determination of structural carbohydrates and acid insoluble lignin was carried out according to NREL protocols (NREL/TP-510-42618).[4] Therefore, dried samples of extractives-free biomass (~0.3 g) were subjected to acid hydrolysis by treating them with sulfuric acid (3 mL, 72% w/w) in a water bath at 30 °C for 2 hours. Then, the solutions were diluted to a concentration of sulfuric acid of 4% w/w and autoclaved at 121 °C for 1 hour. The hydrolysates were

filtered using fritted ceramic funnels (pore size: 4-8 µm) and the acid insoluble lignin content was measured as the weight of the acid insoluble residue minus the ashes. The hydrolysates were then filtered (nylon filters, pore size: 0.2 µm) and analyzed for carbohydrates via High Performance Liquid Chromatography (Agilent 1200 series). A Hi Plex-H column (Biorad) and refractive index detector (RID) were used to determine the concentrations of glucose, xylose, and arabinose at 65 °C using 0.005M $H2_SO_4$ as the mobile phase (eluent) with a flow rate of 0.6 mL min^{-1}. Response factors were determined by calibration with commercial standards.

The equations below summarize the calculations made for the quantification of C5 polysaccharides (xylan and arabinan), C6 polysaccharides (glucan) and acid insoluble lignin (AIL).

$$\mathrm{C5,C6}_{extractives-free}\ (\mathrm{wt\%}) = \frac{\mathrm{C_{anhydro}\ V_{hydrolysate}\ \frac{1\ L}{1000\ mL}}}{m_{DM,extractives-free}} \quad \text{Eq. 1}$$

$$\mathrm{C5,C6}_{as\ received}\ (\mathrm{wt\%}) = \mathrm{C5,C6}_{extractives-free}(1 - \%\mathrm{Extractives}) \quad \text{Eq. 2}$$

$$\mathrm{AIL}_{extractives-free}\ (\mathrm{wt\%}) = \frac{m_{AIL}}{m_{DM,extractives-free}} \quad \text{Eq. 3}$$

$$\mathrm{AIL}_{as\ received}\ (\mathrm{wt\%}) = \mathrm{AIL}_{extractives-free}(1 - \%\mathrm{Extractives}) \quad \text{Eq. 4}$$

Wherein

Subscript “Extractives-free” indicates the content of a component in the extractives-free biomass.

Subscript “As-received” indicates the content of a component in the raw biomass.

$C_{anhydro}$ indicates the concentration of the carbohydrates converted into their polymeric form (glucose in form of glucan, etc.) using an anhydro correction (0.88 for pentoses and 0.90 for hexoses) also corrected for any degradation that may have occurred during the dilute-acid step of the hydrolysis according to methodology well known in the art (e.g. by using a recovery factor calculated from replicates enriched with known concentrations of the carbohydrates analyzed) [g L^{-1}].

$V_{hydrolysate}$ indicates the volume of the hydrolysate [mL].

m_{DM} indicates the amount of dry matter [g].

%Extractives indicates content of extractives in the raw biomass [wt%].

m_{AIL} indicates the amount of acid insoluble lignin [g].

Dithionite-assisted organosolv fractionation and products separation

The dithionite-assisted organosolv fractionation (DAOF) experiments were carried out in a Parr reactor (Parr Instrument Company, Moline, IL, U.S.). The lignocellulosic biomass (3 g) was loaded in the reactor, together with sodium dithionite (1g), 60 mL of n-butanol and 60 mL of milli-Q water. The reactor was sealed, purged with N_2, then pressurized with 30 bar of N_2, introduced at ambient temperature. Subsequently, the mixture was stirred (750 rpm) and the temperature was ramped up to 200 °C, at a rate of ~10 °C min^{-1}. When the set-point temperature was reached, it was maintained constant and the mixture was left to react for a duration of 3 hours.

Afterwards, the reactor was cooled down to room temperature and depressurized. The reactor content was collected and centrifuged (8000 rpm, 10 min) to separate the solid pulp from the liquid fraction. The pulp underwent two consecutive washing cycles, first with n-butanol (40 mL) then with water (40 mL), centrifuging each time to separate the solid and liquid fractions. The washing liquids were added to the liquid fraction from the reaction and filtered under vacuum (glass fiber filter, pore size: 1.6 µm) to eliminate residual solid particles. Subsequently, the liquid was

introduced into a separating funnel and let to rest overnight. Thereafter, an aqueous liquid fraction and an organic liquid fraction were collected and stored at -20 °C for further analysis.
The recovered solid fraction was dried at 60 °C to a constant weight, to remove residual solvent, then it was stored at ambient temperature in view of further analysis.

Analysis of the solid fraction
The dried solid fraction obtained after the organosolv process was analyzed for dry matter and ash, as well as for carbohydrates and acid insoluble lignin, according to the procedures reported above for biomass characterization. Recovery and removal of polysaccharides and lignin in the pulp were calculated with respect to the amount of polysaccharides and lignin in the initial biomass, and with respect to the DM initially introduced, according to the following equations.

$$Recovery_{C5/C6/lignin}\big|_{C5/C6/AIL}\ (\mathrm{wt\%}) = \frac{m_{C5/C6/AIL,pulp}}{m_{C5/C6/AIL,in}} \quad \text{Eq. 5}$$

$$Recovery_{C5/C6/lignin}\big|_{DM}\ (\mathrm{wt\%}) = \frac{m_{C5/C6/lignin,pulp}}{m_{DM,in}} \quad \text{Eq. 6}$$

Wherein
Subscript “pulp” indicates the content of a component in the pulp.
Subscript “in” indicates the content of a component in the raw biomass.
$m_{C5/C6/AIL}$ indicates the amount of C5 or C6 polysaccharides or acid insoluble lignin [g].
m_{DM} indicates the amount of dry matter [g].

Cellulose crystallinity in the solid fraction was determined by subjecting pulp samples to X-ray powder diffraction. Therefore, samples of pulp were milled to obtain a fine powder (particle size ≤ 0.1 mm) and analyzed with a D8 advanced diffractometer, equipped with XE-T detector (Bruker) using a Bragg Brentano geometry. Diffractograms were recorded in a 5 – 80° 2θ range, with an increment of 0.015° (2θ) and an integration time of 0.15 s. The cellulose crystallinity index (CI) was determined for each sample according to Segal’s method.[5] The equation adopted for the calculation of CIs is reported below.

$$CI\ (\%) = \frac{I_{002} - I_{AM}}{I_{002}} \quad \text{Eq. 7}$$

Wherein
I_{002} indicates the maximum intensity of the diffraction at 2θ ~ 22.5°, assigned to the crystalline portion of cellulose and corresponding to the (002) lattice planes.
I_{AM} indicates the intensity of the diffraction at 2θ ~ 18°, assigned to the amorphous portion and corresponding to the minimum intensity between the peaks for the (002) and (101) lattice planes.

Microstructural analysis of the solid fraction was carried out via field emission gun scanning electron microscopy (FEG-SEM). Therefore, solid samples were milled to obtain a fine powder (particle size ≤ 0.1 mm). Specimens were mounted on stubs and coated with a 10 nm gold layer (Cressington sputter 208HR) to create a thin conductive layer, minimizing degradation and drift due to thermal expansion. FEG-SEM analyses were performed in a Jeol FEG-SEM 7600F, operating at 15 keV, with a working distance between 9.5 and 12 mm.

Enzymatic convertibility of the solid fraction was determined by subjecting pulp samples to enzymatic hydrolysis, according to NREL protocol (NREL/TP-510-42629)[6]. Therefore, pulp samples (0.1 g) were introduced in glass tubes and mixed with a citrate buffer (4.72 mL, pH 4.8) and sodium azide (50 µL). The hydrolysis was performed with an enzyme

loading of 15 FPU (Cellic CTec2), at a temperature of 50 °C and samples were shaken at 150 rpm for a duration of 72 hours. Subsequently, the samples were centrifuged to separate residual solids, filtered (nylon filters, pore size: 0.2 µm) and monosaccharides in the liquid were analyzed via High Performance Liquid Chromatography (HPLC), applying the same conditions illustrated above for the analysis of structural carbohydrates in biomass. Conversion yields were calculated with respect to the amount of polysaccharides in the pulp determined via acid hydrolysis, according to the following equation.

$$Conversion_{C5,C6}\ (\text{wt\%}) = \frac{C_{Monosaccharide}}{C_{DM,pulp}\ (C5, C6_{as\ received}/AF)} \quad \text{Eq. 8}$$

Wherein

$C_{Monosaccharide}$ indicates the concentration of monosaccharide determined by HPLC, after enzymatic hydrolysis [g L^{-1}].

$C_{DM,pulp}$ indicates the concentration of dry matter from the pulp [g L^{-1}].

$C5,C6_{as\ received}$ indicates the concentration of polysaccharide in the pulp, determined via acid hydrolysis [wt% DM].

AF indicates the anhydro factor for the conversion of carbohydrates into their polymeric form (glucose in form of glucan, etc.), equal to 0.88 for pentoses and 0.90 for hexoses.

The degree of polymerization (DP) of cellulose in the solid fraction was measured by viscosimetry according to the NF G 06-037 norm. Therefore, samples of pulp (~0.075 g) were dissolved in 15 mL of a 0.5 M cupriethylenediamine solution. The solution was stirred for 2 hours at room temperature. Viscosity data were determined in a UBBELOHDE thermostated capillary tube viscosimeter at 298 K. The DP was determined according to the NF G 06-037 norm.

Analysis of the organic fraction

The organic liquid fraction obtained after the organosolv process was analyzed for dry matter and ash according to the protocols reported above for biomass characterization, then lignin oil was extracted and analyzed following a procedure developed by Van Den Bosch et al.[7] and widely adopted in the literature.[8–13] Therefore, a sample of the organic phase (~5 mL) was dried under nitrogen flow to remove the solvent, then underwent a three-fold liquid-liquid extraction with dichloromethane (6 mL) and water (6 mL), recycling the water phase. The three dichloromethane extracts were mixed, and the solvent was removed by drying under vacuum to determine the weight of lignin oil. A correction was applied to account for the presence of extractives in lignin oil, and the yield of oil was calculated with respect to acid insoluble lignin and with respect to the DM introduced, according to the following equations.

$$Yield_{Lignin\ oil}\big|_{AIL}\ (\text{wt\%}) = \frac{m_{Lignin\ oil} - m_{EtOH\ extractives}}{m_{AIL,in}} \quad \text{Eq. 9}$$

$$Yield_{Lignin\ oil}\big|_{DM}\ (\text{wt\%}) = \frac{m_{Lignin\ oil} - m_{EtOH\ extractives}}{w_{DM,in}} \quad \text{Eq. 10}$$

Wherein

$m_{Lignin\ oil}$ indicates the total amount of lignin oil obtained from the process [g].

$m_{EtOH\ extractives}$ indicates the amount of ethanol extractives present in the biomass initially introduced in the reactor [g].

$m_{AIL,in}$ indicates the amount of acid insoluble lignin present in the biomass initially introduced in the reactor [g].

$m_{DM,in}$ indicates the amount of dry matter present in the biomass initially introduced in the reactor [g].

The molecular weight distribution of the lignin oil was then investigated via gel permeation chromatography (GPC). Therefore, lignin oil was dissolved in tetrahydrofuran to achieve a concentration of about 5 mg mL^{-1} and the solution

was filtered (PTFE filter, pore size: 0.2 µm). GPC analysis was performed at 40 °C, using tetrahydrofuran as a solvent (flowrate: 1 ml min^{-1}) on a Waters E2695 equipped with an Agilent PL Gel column (Mixed E, 3 µm) and a Waters 2998 Photodiode array detector (UV detection at 280 nm). Commercial standards for phenolic monomers and polystyrene standards were employed to create calibration curves.

In order to determine the phenolic monomers composition in the lignin oil, the dichloromethane extract, obtained following a procedure analogous to that reported above, was added with a known amount of 2-isopropyl phenol (internal standard) and analyzed via gas chromatography (GC).

Identification of phenolic monomers was performed using a Thermo Fisher Scientific Trace 1310 equipped with a Rxi-5Sil MS column and an ISQ QD Mass Spectroscopy (MS) detector. The following operating conditions were used: injection temperature of 280 °C, column temperature program: 40 °C (1 min), 10 °C/min to 300 °C (5 min), detection temperature of 310 °C.

Quantification of phenolic monomers was carried out using a Thermo Fisher Scientific Trace GC Ultra equipped with a Rxi-5Sil MS column and a flame ionization detector (FID). The following operating conditions were adopted: injection temperature 280 °C, column temperature program: 40 °C (1 min), 2 °C/min to 150 °C, 5 °C/min to 240 °C, 30 °C/min to 300 °C (15 min), detection temperature of 305 °C. Response factors for the different products were determined by calibration with commercial standards or via calculations based on Effective Carbon Number theory.

The yield of phenolic monomers was calculated with respect to the amount of acid insoluble lignin and with respect to the DM introduced, according to the equations below.

$$Yield_i|_{AIL}\ (\text{wt\%}) = \frac{m_i}{m_{AIL,in}} \qquad \text{Eq. 11}$$

$$Yield_i|_{DM}\ (\text{wt\%}) = \frac{m_i}{m_{DM,in}} \qquad \text{Eq. 12}$$

Wherein

m_i indicates the amount of component *i* obtained from the process, determined via GC-FID analysis [g].

$m_{AIL,in}$ indicates the amount of acid insoluble lignin present in the biomass initially introduced in the reactor [g].

$m_{DM,in}$ indicates the amount of dry matter present in the biomass initially introduced in the reactor [g].

Analysis of organic acids and dehydration products from carbohydrates was carried out by subjecting the organic liquid phase to High Performance Liquid Chromatography. Therefore, a sample of the organic phase was filtered (PTFE filter, pore size: 0.2 µm) and analyzed via High Performance Liquid Chromatography (HPLC), applying the same conditions illustrated above for the analysis of structural carbohydrates in biomass. Peaks identification was based on the comparison of retention times with those of pure standards. Response factors were determined by calibration with commercial standards. The yield of carbohydrate derivatives was calculated with respect to the total amount of polysaccharides in the initial biomass and with respect to the DM introduced, according to the following equations.

$$Yield_i|_{C5+C6}\ (\text{wt\%}) = \frac{m_i}{m_{C5+C6,in}} \qquad \text{Eq. 13}$$

$$Yield_i|_{DM}\ (\text{wt\%}) = \frac{m_i}{m_{DM,in}} \qquad \text{Eq. 14}$$

Wherein

m_i indicates the amount of component *i* obtained from the process, determined via HPLC analysis [g].

$m_{C5+C6,in}$ indicates the total amount of polysaccharides present in the biomass initially introduced in the reactor [g].
$m_{DM,in}$ indicates the amount of dry matter present in the biomass initially introduced in the reactor [g].

Analysis of the aqueous fraction

The analytical procedures applied to characterize the aqueous liquid fraction are analogous to those described for the organic liquid fraction.

Overall mass balance

An overall mass balance for the main biomass components (C5, C6 carbohydrates and lignin) was carried out considering three product streams: a solid fraction, an organic liquid fraction, an aqueous liquid fraction. In addition, a residual "not found" fraction was introduced to account for losses (mainly due to the generation of volatiles). Ash was not taken into consideration in the mass balance, since it only accounts for a marginal fraction of DM in biomass, while most of the ash content in the reaction products derives from the reducing agent. In addition, a correction was applied to account for the presence of non-ash DM originating from the reducing agent, determined via *blank* reactions carried out in absence of biomass. Therefore, the following system of equations (Eq. 15–22) describes the mass balance:

$$C5_S + C5_O + C5_A + C5_{NF} = C5_{in} \quad \text{Eq. 15}$$

$$C6_S + C6_O + C6_A + C6_{NF} = C6_{in} \quad \text{Eq. 16}$$

$$L_S + L_O + L_A + L_{NF} = L_{in} \quad \text{Eq. 17}$$

$$Extr_S + Extr_O + Extr_A + Extr_{NF} = Extr_{in} \quad \text{Eq. 18}$$

$$C5_S + C6_S + L_S + Extr_S + Other_S = Solid_{tot} \quad \text{Eq. 19}$$

$$C5_O + C6_O + L_O + Extr_O + Other_O = Organic_{tot} \quad \text{Eq. 20}$$

$$C5_A + C6_A + L_A + Extr_A + Other_A = Aqueous_{tot} \quad \text{Eq. 21}$$

$$C5_{NF} + C6_{NF} + L_{NF} + Extr_{NF} + Other_{NF} = Residual_{tot} \quad \text{Eq. 22}$$

Wherein
Subscript "S" indicates the amount of a component in the pulp.
Subscript "O" indicates the amount of a component in the organic liquid fraction.
Subscript "A" indicates the amount of a component in the aqueous liquid fraction.
Subscript "NF" indicates the amount of a component in the "not found" fraction.
Subscript "in" indicates the amount of a component in the biomass initially introduced in the reactor.
C5 indicates the amount of xylan and arabinan [g].
C6 indicates the amount of glucan [g].

L indicates the amount of acid-insoluble lignin [g].
Extr indicates the amount of extractives [g].
Other indicates the amount of biomass components that do not originate from C5, C6, lignin or extractives [g].
$Solid_{tot}$ indicates the total amount of biomass components in the solid fraction [g].
$Organic_{tot}$ indicates the total amount of biomass components in the organic liquid fraction [g].
$Aqueous_{tot}$ indicates the total amount of biomass components in the aqueous liquid fraction [g].
$Residual_{tot}$ indicates the total amount of biomass components that are not recovered in the solid or liquid product streams [g].

Variables that can be measured include: $C5_S$, $C5_{in}$, $C6_S$, $C6_{in}$, L_S, L_{in}, $Extr_{in}$, $Solid_{tot}$, $Organic_{tot}$, $Aqueous_{tot}$, $Residual_{tot}$.
Unknown variables include: $C5_O$, $C5_A$, $C5_{NF}$, $C6_O$, $C6_A$, $C6_{NF}$, L_O, L_A, L_{NF}, $Extr_S$, $Extr_O$, $Extr_A$, $Extr_{NF}$, $Other_S$, $Other_O$, $Other_A$, $Other_{NF}$.
Thus, the system is unsaturated. In order to solve the mass balance, the following assumptions were introduced:

- Water- and ethanol-soluble extractives are entirely recovered in the aqueous phase and in the organic phase, respectively.
- Lignin contributes marginally to the formation of volatile compounds (L_{NF} is negligible).
- The components deriving from C5 and C6 in the organic and aqueous fractions consist either of condensed derivatives (humins) incorporated in lignin oil, or non-condensed derivatives (saccharides, polyols and organic acids).
- The amount of condensed derivatives from C5 (and C6) in lignin oil is inversely proportional to the amount of C5 (and C6) in the solid fraction.
- The amount of non-condensed derivatives from C5 (and C6) in the organic or aqueous fraction is inversely proportional to the amount of C5 (and C6) in the solid fraction.
- Solubilized lignin and humins migrate to the organic or the aqueous phase proportionally to the amount of lignin oil contained in that phase.

Leading to the additional equations (Eq. 23 – 38).

$$Extr_A = Extr_{in,H_2O} \quad \text{Eq. 23}$$

$$Extr_O = Extr_{in,EtOH} \quad \text{Eq. 24}$$

$$Extr_S = 0 \quad \text{Eq. 25}$$

$$L_{NF} = 0 \quad \text{Eq. 26}$$

$$C5_O = C5_{OL} + C5_{OC} \quad \text{Eq. 27}$$

$$C6_O = C6_{OL} + C6_{OC} \quad \text{Eq. 28}$$

$$C6_{OL} = \frac{C6_{in} - C6_S}{(C6_{in} - C6_S) + (C5_{in} - C5_S)} (C5_{OL} + C6_{OL}) \quad \text{Eq. 29}$$

$$C5_{OC} + C6_{OC} = CarbDer_O \quad \text{Eq. 30}$$

$$C6_{OC} = \frac{C6_{in} - C6_S}{(C6_{in} - C6_S) + (C5_{in} - C5_S)} (C5_{OC} + C6_{OC}) \quad \text{Eq. 31}$$

$$C5_A = C5_{AL} + C5_{AC} \quad \text{Eq. 32}$$

$$C6_A = C6_{AL} + C6_{AC} \quad \text{Eq. 33}$$

$$C6_{AL} = \frac{C6_{in} - C6_S}{(C6_{in} - C6_S) + (C5_{in} - C5_S)} (C5_{AL} + C6_{AL}) \quad \text{Eq. 34}$$

$$C5_{AL} + C6_{AL} = \frac{LigninOil_A}{LigninOil_O} (C5_{OL} + C6_{OL}) \quad \text{Eq. 35}$$

$$L_A = \frac{LigninOil_A}{LigninOil_O} (L_O) \quad \text{Eq. 36}$$

$$C5_{AC} + C6_{AC} = CarbDer_A \quad \text{Eq. 37}$$

$$C6_{AC} = \frac{C6_{in} - C6_S}{(C6_{in} - C6_S) + (C5_{in} - C5_S)} (C5_{AC} + C6_{AC}) \quad \text{Eq. 38}$$

Wherein

Subscript “OL” indicates the amount of a component in the organic liquid fraction, contributing to lignin oil.

Subscript “OC” indicates the amount of a component in the organic liquid fraction, contributing to non-condensed carbohydrate derivatives.

Subscript “AL” indicates the amount of a component in the aqueous liquid fraction, contributing to lignin oil.

Subscript “AC” indicates the amount of a component in the aqueous liquid fraction, contributing to non-condensed carbohydrate derivatives.

$Extr_{in,H_2O}$ indicates the amount of water-soluble extractives in biomass [g].

$Extr_{in,EtOH}$ indicates the amount of ethanol-soluble extractives in biomass [g].

CarbDer indicates the amount of non-condensed C5, C6 derivatives [g].

LigninOil indicates the amount of lignin oil [g].

Variables that can be measured include: $CarbDer_O$, $CarbDer_A$, $LigninOil_O$, $LigninOil_A$.

Unknown variables include: $C5_{OL}$, $C5_{OC}$, $C6_{OL}$, $C6_{OC}$, $C5_{AL}$, $C5_{AC}$, $C6_{OL}$, $C6_{AC}$.

Overall, considering Eq. 15–38, the system of equations has one degree of freedom. Instead of making further assumptions to saturate the system, the two following inequalities were considered, thanks to which it was possible to determine upper and lower limits for all the unknown variables.

$$Other_O \geq 0 \quad \text{Eq. 39}$$

$$Other_{NF} \geq 0 \qquad \text{Eq. 40}$$

Assessment of butanol recovery

The assessment of the recovery of the butanol co-solvent in the liquid fractions was performed by measuring the butanol content of each fraction via High Performance Liquid Chromatography, following a procedure analogous to that described above for the analysis of organic acids and dehydration products from carbohydrates. The recovery of butanol in each liquid fraction was calculated with respect to the total amount of butanol utilized during the process (for the reaction and for washing the isolated pulp), according to the following equation.

$$Recovery_{Butanol}|_{O/A}\ (\text{wt\%}) = \frac{C_{Butanol,O/A}\ V_{O/A}\ \frac{1\ \text{L}}{1000\ \text{mL}}}{m_{Butanol,in} + m_{Butanol,wash}} \qquad \text{Eq. 41}$$

Wherein

$C_{Butanol,O/A}$ indicates the concentration of butanol measured in the organic or aqueous fraction [g L^{-1}].

$V_{O/A}$ indicates the volume of organic or aqueous fraction [mL].

$m_{Butanol,in}$ indicates the amount of butanol employed for the fractionation [g].

$m_{Butanol,wash}$ indicates the amount of butanol employed for washing the isolated pulp [g].

Analysis of dithionite derivatives

In order to inspect the presence of derivatives from dithionite in the isolated fractions, the sulfur content in each fraction was determined via mineralization followed by Inductively Coupled Plasma Atomic Emission Spectroscopy (ICP-AES). Therefore, a sample of the solid fraction (~20 mg) was mixed with 2 mL of HNO_3 (69%) and 1 mL of H_2O_2 (30%) and the mixture was left to react for 10 minutes. Then, the mixture was dried at 100 °C. The obtained residue was redispersed in 2 mL of HNO_3 before being diluted with 25 mL water for the analysis. An analogous procedure was followed for the mineralization of the organic fraction (a sample of 0.5 mL was used in this case). Sample preparation for the aqueous fraction consisted simply in the dilution of a sample in HNO_3 (8%). ICP-AES analyses were performed using a Thermo iCAP 6500 Duo spectrometer. The system was calibrated using pure sulfur standard solutions. The recovery of sulfur in each fraction was calculated with respect to the total amount of sulfur introduced in the process, according to the following equation.

$$Recovery_{Sulfur}\big|_{S/O/A}\ (\%) = \frac{C_{Sulfur,S/O/A}\ \frac{1\ \text{g}}{1000\ \text{mg}}\ m_{S/O/A}\ \frac{1\ \text{kg}}{1000\ \text{g}}}{\frac{N_{Sulfur}\ M_{Sulfur}}{M_{Na_2S_2O_4}}\ m_{Na_2S_2O_4,in}} \qquad \text{Eq. 42}$$

Wherein

$C_{Sulfur,S/O/A}$ indicates the concentration of sulfur measured in the solid, organic or aqueous fraction [mg kg^{-1}].

$m_{Sulfur,S/O/A}$ indicates the amount of solid, organic or aqueous fraction obtained from the process [g].

N_{Sulfur} indicates the number of sulfur atoms in sodium dithionite.

M_{Sulfur} indicates the atomic mass of sulfur [g mol^{-1}].

$M_{Na_2S_2O_4}$ indicates the molecular weight of sodium dithionite [g mol^{-1}].

$m_{Na_2S_2O_4,in}$ indicates the amount of sodium dithionite introduced in the process [g].

Milled wood lignin: preparation and organosolv treatment

Isolation of milled wood lignin (MWL) was performed following a procedure that is well-established in the literature.[14] Therefore, water- and ethanol-soluble extractives were removed from biomass according to the methods reported above, then the extracted biomass was milled at 800 rpm for 12 hours (with pauses of 5 minutes every 5 minutes of

milling to avoid overheating) using a Retsch Emax ball mill. Subsequently, the milled wood was dispersed in an aqueous solution of dioxane (96% v/v) at a concentration of 40 g/L and stirred at ambient temperature for 24 hours. Thereafter, the suspension was centrifuged (8000 rpm, 10 min) and the solid residue was redispersed in fresh aqueous dioxane under stirring for an additional 24 hours. The extracts were combined and dried under vacuum to yield crude MWL. This intermediate product was dissolved in an aqueous solution of acetic acid (90%) at a concentration of 50 g/L. Afterwards, lignin was precipitated by dropwise addition of the acetic acid solution to water (220 mL of water per gram of crude MWL), centrifuged (8000 rpm, 10 min) and air dried to a constant weight. The isolated lignin was refined further by dissolving it in a solution of 1,2-dichloroethane and ethanol (2:1 v/v) at a concentration of 50 g/L, which was then centrifuged to remove residual solids. Lignin was precipitated once again by dropwise addition of the 1,2-dichloroethane – ethanol solution to anhydrous ethyl ether (230 mL of ether per gram of lignin). After centrifugation (8000 rpm, 10 min), the isolated MWL was washed three times with fresh ether, centrifuging after every washing step, and, finally, air dried to a constant weight to yield purified MWL.

MWL was subjected to organosolv treatment performed as described above. Therefore, 0.63 g of MWL were introduced in the reactor, together with 1 g of sodium dithionite, 60 mL of n-butanol and 60 mL of milli-Q water. The reaction was carried out at a temperature of 200 °C, following the procedure illustrated above. The subsequent product separation and analysis was conducted as previously described. For the reaction with MWL and formic acid, 0.63 g of MWL and 0.11 g of formic acid were subjected to an identical process.

Reactions with model compounds

The reactions with model compounds were carried out using the same equipment and an analogous procedure as that presented for organosolv fractionation experiments. Therefore, a weighed amount of the model compound (0.02 g for 2-phenoxy-1-phenylethanol and 0.05 g for 4-propenyl guaiacol and 3-phenyl-1-propenol) was introduced in the reactor, together with 60 mL of n-butanol, 60 mL of milli-Q water and the selected amount of sodium dithionite (between 0 and 12 molar equivalents). The reaction was performed following the procedure described above. For the treatment of 2-phenoxy-1-phenylethanol, a higher reaction temperature of 250 °C was adopted, to promote the cleavage of the dimer.[15] Subsequently, the reactor content was collected, transferred into a separating funnel and let to rest overnight. Thereafter, the aqueous liquid fraction and the organic liquid fraction were collected and stored at -20 °C for further analysis.

The reaction products were then analyzed via GPC and GC-MS/FID following the procedures reported above.

The C-based yields of phenolic monomers were calculated according to the equation below.

$$Yield_i\ (\mathrm{C\%}) = \frac{m_i\ \frac{C_i\ M_C}{MW_i}}{m_{Model}\ \frac{C_{Model}\ M_C}{MW_{Model}}} \qquad \text{Eq. 43}$$

Wherein

m_i indicates the amount of component *i* obtained from the process, determined via GC analysis [g].

C_i indicates the number of carbon atoms in the molecule of component *i*.

M_c indicates the atomic mass of carbon [g mol^{-1}].

MW_i indicates the molecular weight of component *i* [g mol^{-1}].

m_{Model} indicates the amount of model compound introduced in the reactor [g].

C_{Model} indicates the number of carbon atoms in the molecule of the model compound.

MW_{Model} indicates the molecular weight of the model compound [g mol^{-1}].

Simplified economic assessment of monophenolics production via dithionite-assisted organosolv fractionation

Dithionite-assisted organosolv fractionation was shown to enhance depolymerization of lignin from lignocellulosic biomass compared to classical organosolv treatment. With the process configuration examined in this work, about 8.5 grams of sodium dithionite are consumed per gram of phenolic monomers produced. Assuming the process is scalable and considering a market price for sodium dithionite of about 600 €/ton (estimated from industrial suppliers online), the cost of dithionite per ton of phenolic monomers produced would be of about 5100 €. Even though this represents a quite large operative cost, the market values reported in the literature for phenolic monomers range between ~2000 €/ton and ~12000 €/ton,[7,16] suggesting that dithionite-assisted organosolv fractionation can potentially represent an economically viable alternative to conventional pulping processes.

Figures

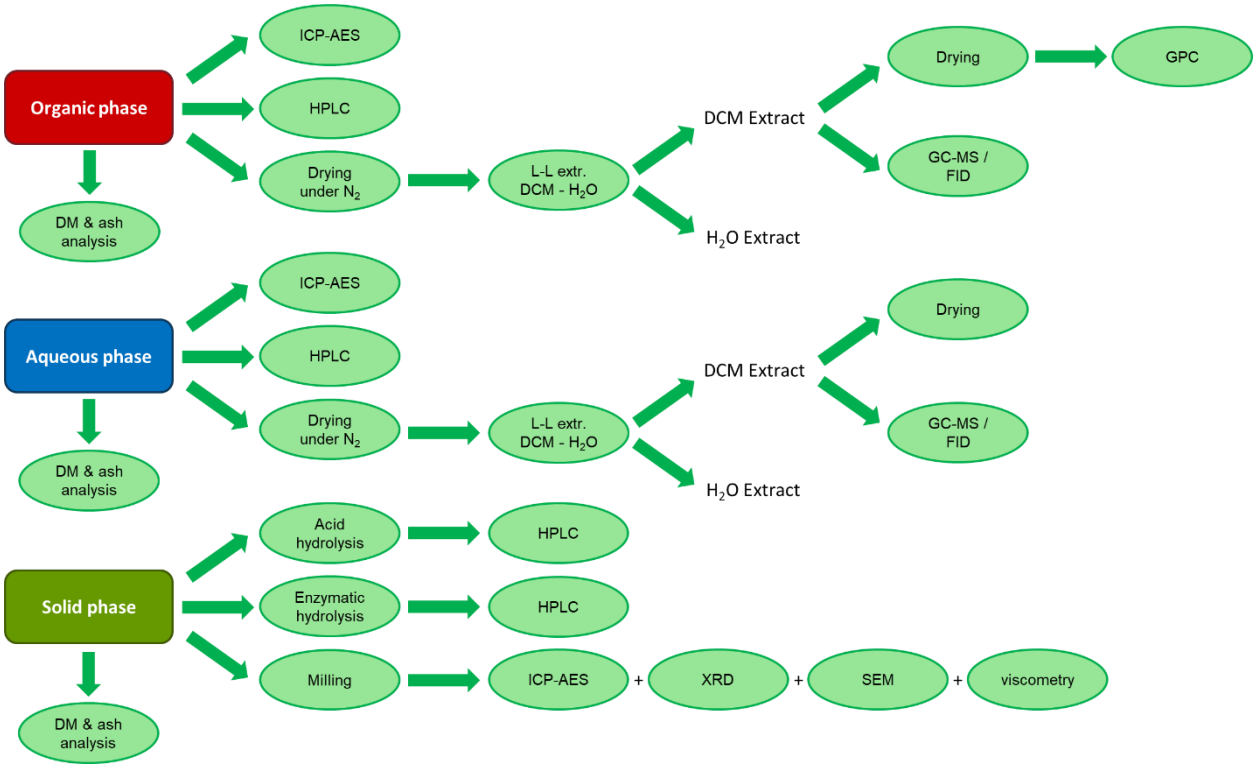


Figure S1 Schematic representation of the analytical procedures adopted for the characterization of the output streams obtained from biomass fractionation.

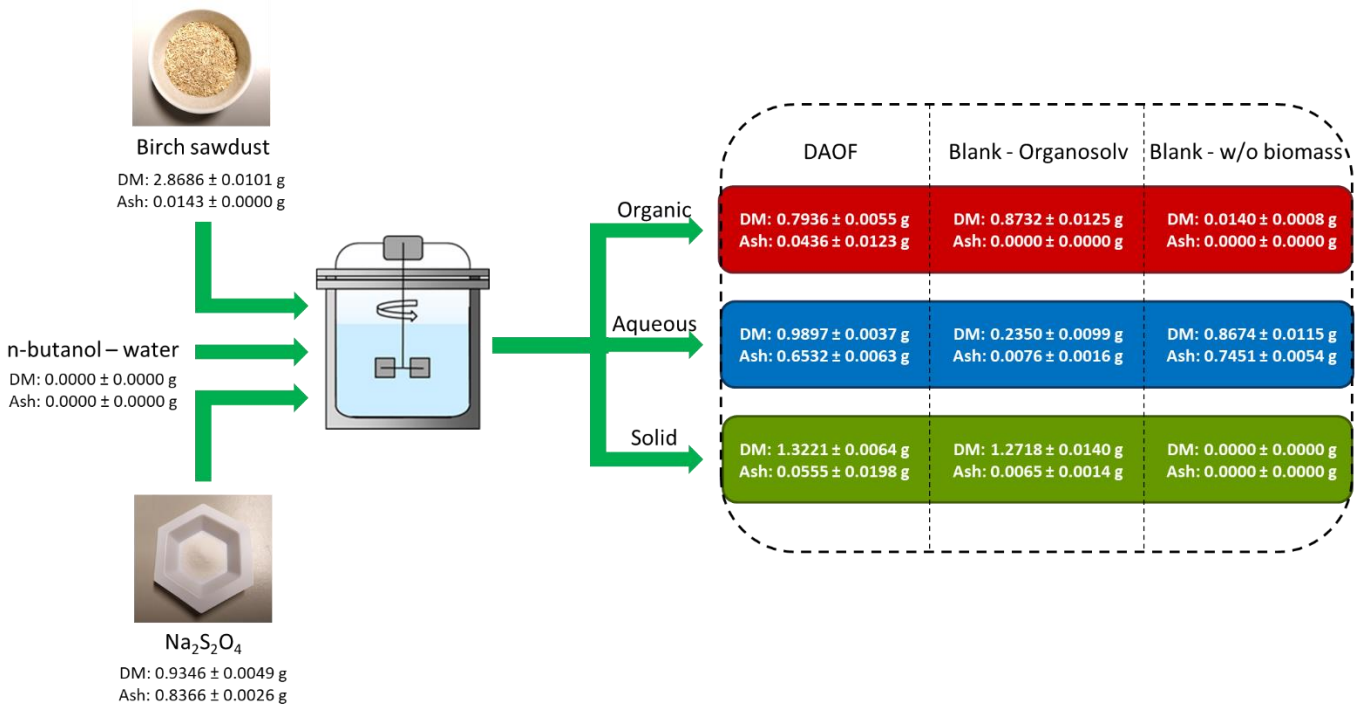


Figure S2 DM and Ash contents measured in each process stream for the dithionite-assisted organosolv fractionation (DAOF) of birch sawdust, for the organosolv treatment of birch sawdust (Blank - Organosolv), and for a reaction performed without biomass (Blank - w/o biomass). The reactions were carried out treating biomass (3 g, particle size < 2 mm) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature), for a duration of 3 hours.

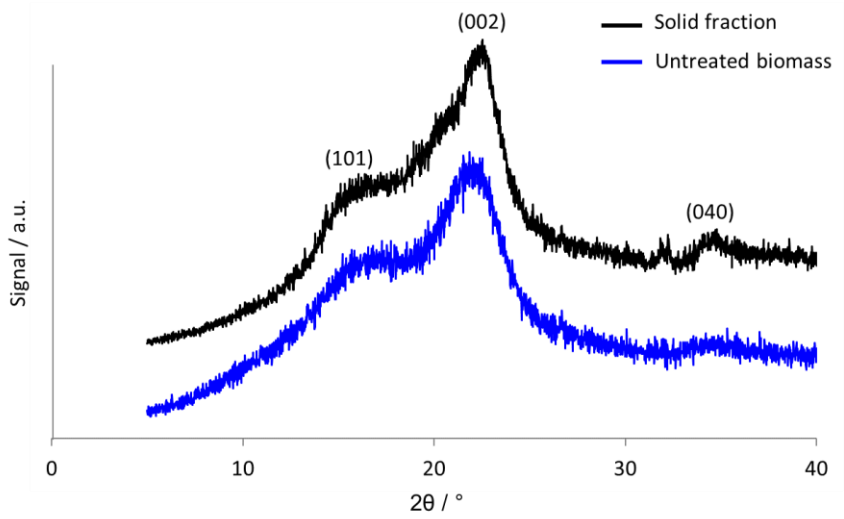


Figure S3 XRD profiles obtained for untreated birch sawdust and for the solid fraction isolated after treating birch sawdust (3 g, particle size < 2 mm) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature), in the presence of $Na_2S_2O_4$ (1 g), for a duration of 3 hours.

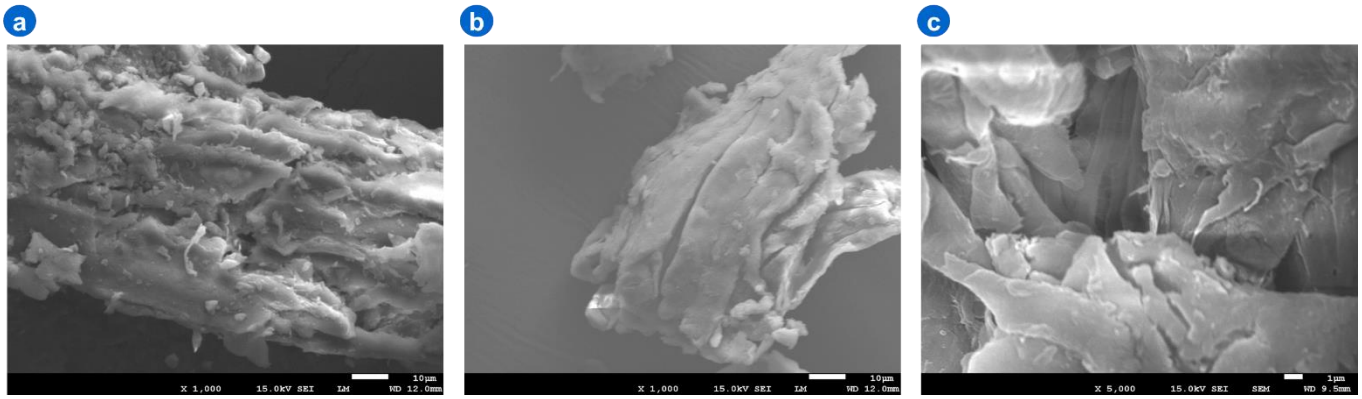


Figure S4 SEM images obtained for untreated birch sawdust (a) and for the solid fraction isolated after treating birch sawdust (3 g, particle size < 2 mm) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature), in the presence of $Na_2S_2O_4$ (1 g), for a duration of 3 hours (b and c). (b) Fiber cells bundles. (c) Detail of fiber cells.

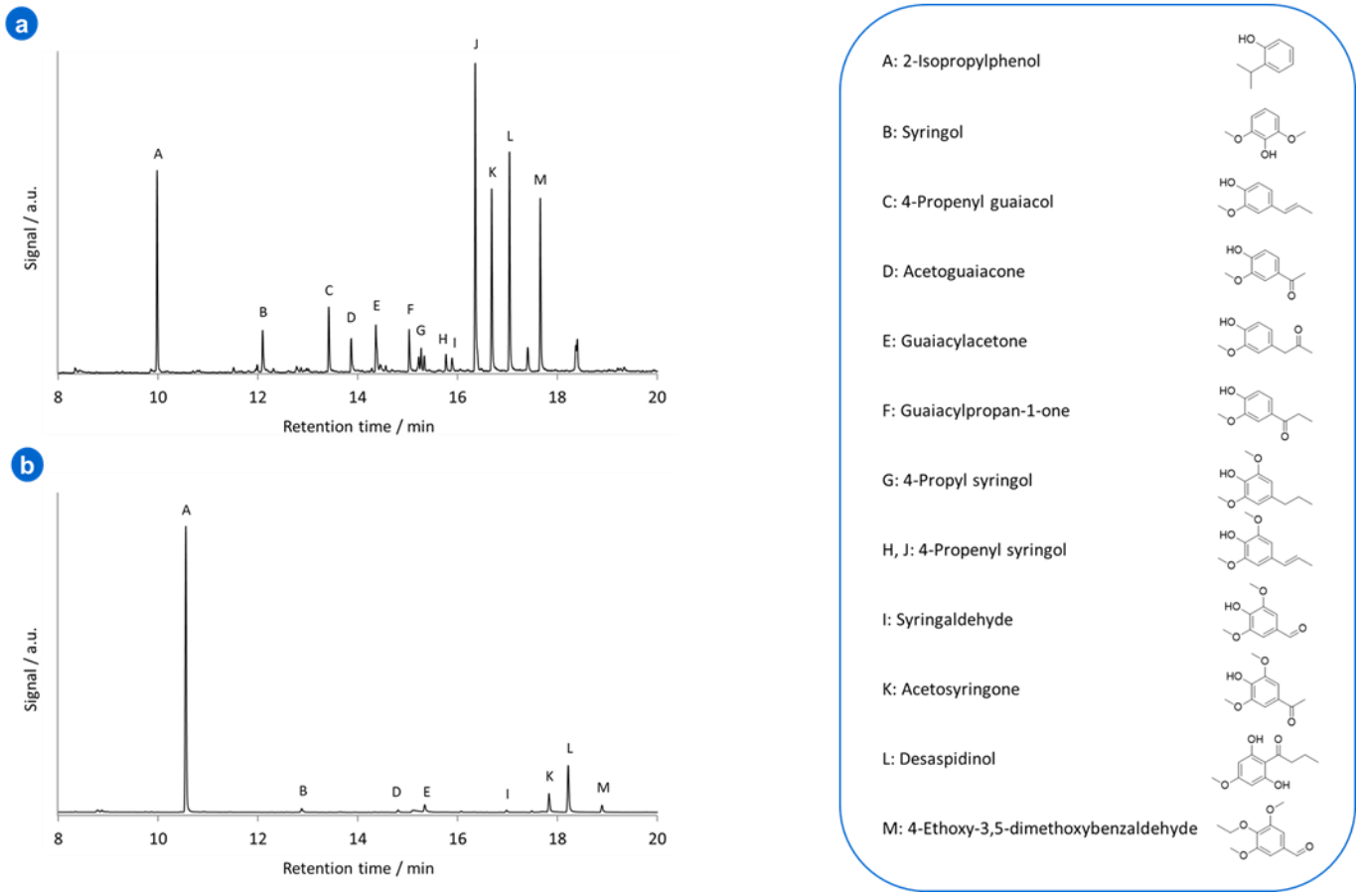


Figure S5 Gas -chromatograms of the lignin oil isolated from the organic fraction (a) and aqueous fraction (b), obtained after treating birch sawdust (3 g, particle size < 2 mm) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature), in the presence of $Na_2S_2O_4$ (1 g), for a duration of 3 hours.

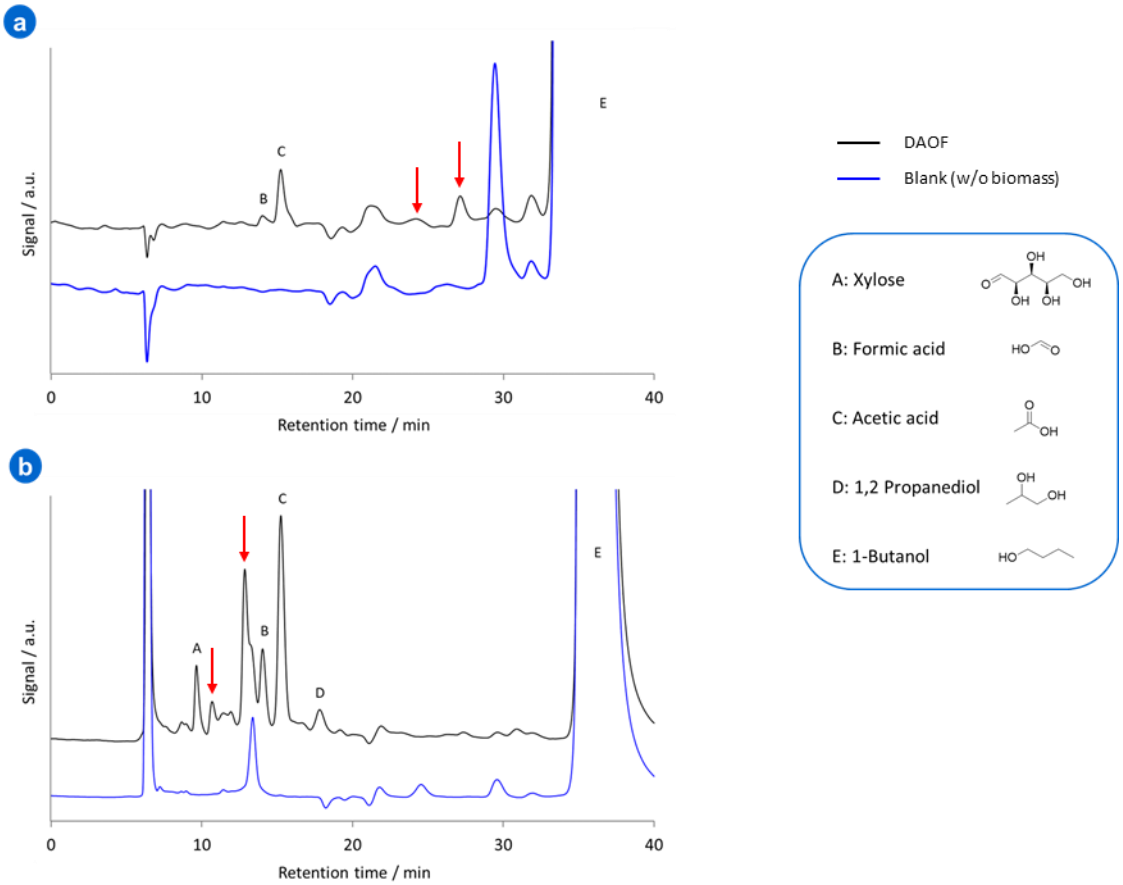


Figure S6 HPLC chromatograms of the organic fraction (a) and aqueous fraction (b), obtained after treating birch sawdust (3 g, particle size < 2 mm) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature), in the presence of 1 g of $Na_2S_2O_4$ (1 g), for a duration of 3 hours. Chromatograms obtained for blank reactions (without biomass), at identical conditions were added for comparison. Red arrows in the chromatograms point toward peaks corresponding to components that originate from biomass, but could not be identified.

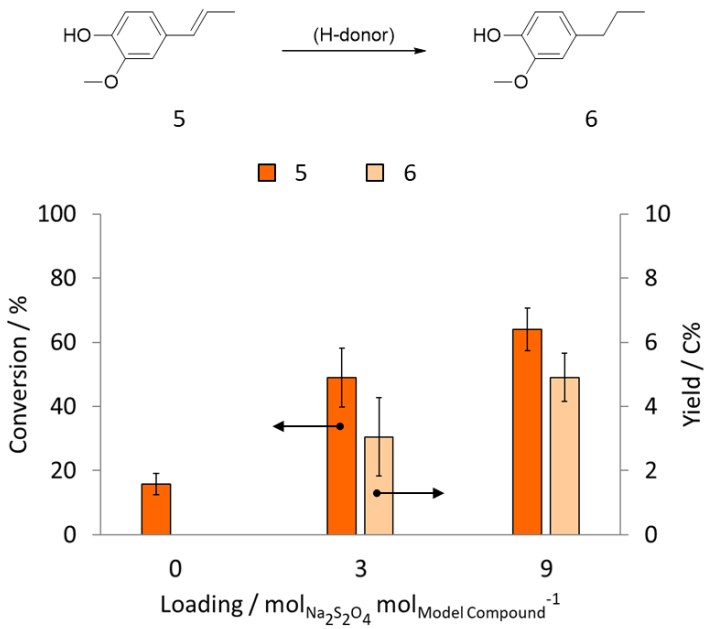


Figure S7 Effect of $Na_2S_2O_4$ on the conversion of 4-propenyl guaiacol and on the yields of monomeric products obtained from the reaction of 4-propenyl guaiacol (0.05 g) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature), for a duration of 3 hours. The reactions were performed in duplicates. The arrows in the figure indicate the reference axis.

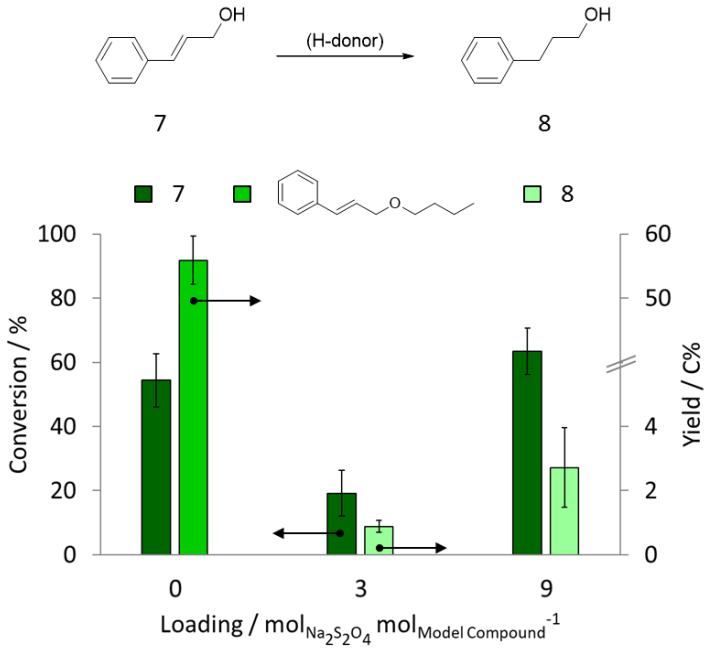


Figure S8 Effect of $Na_2S_2O_4$ on the conversion of the 3-phenyl-1-propenol and on the yields of monomeric products obtained from the reaction of 3-phenyl-1-propenol (0.05 g) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature), for a duration of 3 hours. The reactions were performed in duplicates. The arrows in the figure indicate the reference axis.

Figure S9 Parr batch reactor used for the experimentation.

Figure S10 Organic and aqueous liquid fractions (left), lignin oil (middle) and solid fraction (right) obtained for the dithionite-assisted organosolv fractionation of birch wood.

```
Solve[C5s + C5o + C5a + C5nf == C5in &&
  C6s + C6o + C6a + C6nf == C6in &&
  Ls + Lo + La + Lnf == Lin &&
  Extrs + Extro + Extra + Extrnf == Extrin &&

  C5s + C6s + Ls + Extrs + Others == Solidtot &&
  C5o + C6o + Lo + Extro + Othero == Organictot &&
  C5a + C6a + La + Extra + Othera == Aqueoustot &&
  C5nf + C6nf + Lnf + Extrnf + Othernf == Residualtot &&

  C5o == C5ol + C5oc &&
  C6o == C6ol + C6oc &&

  C6ol == ((C6in - C6s) / ((C6in - C6s) + (C5in - C5s))) * (C5ol + C6ol) &&

  C5oc + C6oc == CarbDero &&
  C6oc == ((C6in - C6s) / ((C6in - C6s) + (C5in - C5s))) * (C6oc + C5oc) &&

  C5a == C5al + C5ac &&
  C6a == C6al + C6ac &&

  C6al == ((C6in - C6s) / ((C6in - C6s) + (C5in - C5s))) * (C6al + C5al) &&
  (C5al + C6al) == LigninOila / LigninOilo * (C5ol + C6ol) &&
  La == LigninOila / LigninOilo * (Lo) &&

  C5ac + C6ac == CarbDera &&
  C6ac == ((C6in - C6s) / ((C6in - C6s) + (C5in - C5s))) * (C6ac + C5ac),
 {C5o, C5a, C5nf, C6o, C6a, C6nf, Lo, La, Extrnf, Others, Othero, Othera, Othernf, C5ol, C5oc, C6ol, C6oc, C5al, C5ac, C6al, C6ac}]
```

Measured variables

```
C5in = 21.8
C6in = 36.6
Lin = 22.0
Extrin = 8.2

Solidtot = 44.1
Organictot = 26.6
Aqueoustot = 17.6
Residualtot = 11.2

C5s = 4.0
C6s = 33.5
Ls = 6.2
Extrs = 0.0
Extro = 2.3
Extra = 5.9

LigninOilo = 20.5
LigninOila = 0.7
CarbDero = 0.1
CarbDera = 4.4

Lnf = 0.0
```

```
{{C5a → 3.74446 + 0.0341463 C5o, C5nf → 14.0555 - 1.03415 C5o, C6o → 0. + 0.174157 C5o, C6a → 0.652125 + 0.00594683 C5o, C6nf → 2.44787 - 0.180104 C5o, Lo → 15.2783, La → 0.521698,
  Extrnf → 0., Others → 0.4, Othero → 9.0217 - 1.17416 C5o, Othera → 6.78172 - 0.0400932 C5o, Othernf → -5.30341 + 1.21425 C5o, C5ol → -0.0851675 + 1. C5o, C5oc → 0.0851675,
  C6ol → -0.0148325 + 0.174157 C5o, C6oc → 0.0148325, C5al → -0.00290816 + 0.0341463 C5o, C5ac → 3.74737, C6al → -0.000506477 + 0.00594683 C5o, C6ac → 0.652632}}
```

```
Reduce[9.021698113207547 - 1.1741573033707866 * C5o ≥ 0 &&
  -5.303414634146343 + 1.2142504795834475 * C5o ≥ 0,
 {C5o}]
```

```
4.36764 ≤ C5o ≤ 7.68355
```

```
C5oset = (7.683551503114562 + 4.3676446691342425) / 2
```

```
6.0256
```

```
Solve[C5a == 3.744460263741394 + 0.03414634146341463 * C5oset &&
  C5nf == 14.055539736258606 - 1.0341463414634147 * C5oset &&
  C6o == 0.17415730337078653 * C5oset &&
  C6a == 0.6521251021122652 + 0.005946834749246371 * C5oset &&
  C6nf == 2.4478748978877363 - 0.1801041381200329 * C5oset &&
  Othero == 9.021698113207547 - 1.1741573033707866 * C5oset &&
  Othera == 6.781716520938795 - 0.040093176212661 * C5oset &&
  Othernf == -5.303414634146343 + 1.2142504795834475 * C5oset &&
  C5ol == -0.08516746411483254 + C5oset &&
  C6ol == -0.014832535885167461 + 0.17415730337078653 * C5oset &&
  C5al == -0.0029081573112381843 + 0.03414634146341463 * C5oset &&
  C6al == -0.000506476835103279 + 0.005946834749246366 * C5oset,
 {C5a, C5nf, C6o, C6a, C6nf, Othero, Othera, Othernf, C5ol, C6ol, C5al, C6al}]
```

```
{{C5a → 3.95021, C5nf → 7.82419, C6o → 1.0494, C6a → 0.687958, C6nf → 1.36264,
  Othero → 1.9467, Othera → 6.54013, Othernf → 2.01317, C5ol → 5.94043, C6ol → 1.03457, C5al → 0.202844, C6al → 0.0353268}}
```

Figure S11 Solution of the equations describing the overall mass balance for the treatment of birch sawdust (3 g, particle size < 2 mm) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature), in the presence of $Na_2S_2O_4$ (1 g), for a duration of 3 hours. Equations and inequalities were solved using Wolfram Mathematica 11.2.

Tables

Table S1 Compositional characterization of birch sawdust.

Component	Content (wt%)[a]
Lignin	
Acid insoluble lignin	22.0 ± 0.2
C5	
Xylan	21.4 ± 1.0
Arabinan	0.4 ± 0.1
C6	
Glucan	36.6 ± 0.3
Extractives	
Ethanol-soluble	2.3 ± 0.1
Water-soluble	5.9 ± 0.1
Ash	0.5 ± 0.0
Other	10.9 ± 1.6

[a] The contents of the different components are expressed as percentages of dry matter. Four replicates were performed for each analysis.

Table S2 Characterization of the solid and liquid fractions obtained from the treatment of birch sawdust (3 g, particle size < 2 mm) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N2 (introduced at ambient temperature), for a duration of 3 hours. No $Na_2S_2O_4$ was added to the mixture. The reaction was carried out in triplicates.

Solid fraction	Recovery (wt%)	C5	15.5 ± 1.1
		C6	92.5 ± 3.0
		Lignin[a]	30.7 ± 8.5
	Conversion (%)	Glucan	87.5 ± 3.9
		Xylan	99.0 ± 4.0
	CI (%)		52.5 ± 1.4
Organic fraction	Yield (wt%)	Lignin oil[b]	95.2 ± 1.9
		Monophenolics[b]	3.4 ± 0.6
		C5, C6 derivatives[c]	0.0 ± 0.0
Aqueous fraction	Yield (wt%)	Lignin oil[b]	4.1 ± 0.4
		Monophenolics[b]	0.1 ± 0.0
		C5, C6 derivatives[c]	3.4 ± 0.5

[a] Acid-insoluble lignin

[b] Expressed with respect to the weight of acid-insoluble lignin contained in the initial biomass.

[c] Non-condensed carbohydrate derivatives, expressed with respect to the total weight of polysaccharides contained in the initial biomass.

Table S3 Values of cellulose degree of polymerization (DP), determined by viscosimetry for the solid fractions obtained from the treatment of birch sawdust (3 g, particle size < 2 mm) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature),) for a duration of 3 hours, in the presence of 1 g of $Na_2S_2O_4$ (DAOF), or in the absence of $Na_2S_2O_4$ (Blank). The DP of Avicel cellulose was measured as a reference standard. Pulp samples were analyzed in duplicates.

Experiment	Cellulose DP (AGU)
DAOF[a]	780.0 ± 23.0
Blank	227.0 ± 2.7
Avicel cellulose	150.0 ± 0.1

[a]A small amount of solid residue was observed which did not dissolve in cupriethylenediamine, likely related to structural modification of the pulp triggered by dithionite (e.g. incorporation of sulfur). This phenomenon likely explains the larger error in the measurement.

Table S4 Phenolic monomer composition of the lignin oil isolated from the organic and the aqueous fractions obtained after treating birch sawdust (3 g, particle size < 2 mm) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature), in the presence of $Na_2S_2O_4$ (1 g), for a duration of 3 hours. The reaction was carried out in triplicates.

Component	Organic fraction Yield (wt%)[a]	Aqueous fraction Yield (wt%)[a]
Syringol	0.6 ± 0.0	0.0 ± 0.0
4-Propenyl guaiacol	0.8 ± 0.1	-
Acetoguaiacone	0.5 ± 0.0	0.0 ± 0.0
Guaiacylacetone	0.5 ± 0.0	0.0 ± 0.0
Guaiacylpropan-1-one	0.5 ± 0.0	-
4-Propyl syringol	0.3 ± 0.0	-
Syringaldehyde	0.5 ± 0.0	0.0 ± 0.0
4-Propenyl syringol	4.3 ± 0.9	-
Acetosyringone	2.7 ± 0.2	0.1 ± 0.0
Desaspidinol	3.2 ± 0.5	0.2 ± 0.0
4-Ethoxy-3,5-dimethoxybenzaldehyde	4.3 ± 0.1	0.1 ± 0.0
Total	18.1 ± 1.7	0.5 ± 0.1

[a] Expressed with respect to the weight of acid-insoluble lignin contained in the initial biomass.

Table S5 Phenolic monomer composition of the lignin oil isolated from the organic and the aqueous fractions obtained after treating birch sawdust (3 g, particle size < 2 mm) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature), for a duration of 3 hours. No $Na_2S_2O_4$ was added to the mixture. The reaction was carried out in triplicates.

Component	Organic fraction Yield (wt%)[a]	Aqueous fraction Yield (wt%)[a]
Guaiacol	0.1 ± 0.0	-
Syringol	0.0 ± 0.0	
Methyl syringol	0.3 ± 0.0	-
Syringaldehyde	1.0 ± 0.3	0.0 ± 0.0
4-Propenyl syringol	0.9 ± 0.0	-
Acetosyringone	0.4 ± 0.0	-
Desaspidinol	0.2 ± 0.1	0.0 ± 0.0
4-Ethoxy-3,5-dimethoxybenzaldehyde	0.1 ± 0.0	-
Sinapyl aldehyde	0.2 ± 0.0	-
Sinapyl alcohol	0.4 ± 0.1	-
Total	3.6 ± 0.6	0.1 ± 0.0

[a] Expressed with respect to the weight of acid-insoluble lignin contained in the initial biomass.

Table S6 Characterization of biomass derivatives in the output streams obtained from the treatment of birch sawdust (3 g, particle size < 2 mm) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature),) for a duration of 3 hours, in the presence of 1 g of $Na_2S_2O_4$ (DAOF), or in the absence of $Na_2S_2O_4$ (Blank). The reactions were carried out in triplicates. The ash fraction was not taken into consideration, since it only accounts for a marginal fraction of DM in biomass (0.5%), while most of the ash content in the reaction products derives from $Na_2S_2O_4$.

Solid fraction (wt% DM_{in})		**DAOF**	**Blank**
Non-volatile[a]	Non-ash DM[b]	44.1 ± 0.9	44.1 ± 0.6
	C6	33.5 ± 0.4	33.8 ± 1.0
	C5	4.0 ± 0.7	3.3 ± 0.2
	Lignin[c]	6.2 ± 0.6	6.8 ± 1.9
	Non-ash other	0.4 ± 0.1	0.2 ± 0.4
Total		44.1 ± 0.9	44.1 ± 0.6
Organic fraction (wt% DM_{in})		**DAOF**	**Blank**
Non-volatile[a]	Non-ash DM[b]	25.6 ± 0.6	30.4 ± 0.4
	Lignin monomers	4.0 ± 0.1	0.8 ± 0.1
	Lignin oligomers[d]	16.5 ± 1.6	20.6 ± 0.4
	Extractives	2.3 ± 0.1	2.3 ± 0.1
	Non-ash other	2.9 ± 1.3	7.2 ± 0.9
Volatile[e]	Formic acid	0.1 ± 0.0	0.0 ± 0.0
	Acetic acid	0.9 ± 0.0	0.4 ± 0.0
Total		26.6 ± 0.7	30.9 ± 0.5
Aqueous fraction (wt% DM_{in})		**DAOF**	**Blank**
Non-volatile[a]	Non-ash DM[b]	7.5 ± 0.9	7.9 ± 0.4
	Lignin monomers	0.1 ± 0.0	0.0 ± 0.0
	Lignin oligomers[d]	0.6 ± 0.1	0.9 ± 0.1
	Xylose	0.1 ± 0.0	1.2 ± 0.1
	1,2 Propanediol	0.7 ± 0.1	0.4 ± 0.0
	Extractives	5.9 ± 0.1	5.9 ± 0.1
	Non-ash other	0.1 ± 0.6	-0.4 ± 0.7
Volatile[e]	Formic acid	3.7 ± 0.2	0.5 ± 0.2
	Acetic acid	6.5 ± 0.3	3.1 ± 0.1
Total		17.6 ± 0.7	11.4 ± 0.7
Not found (wt% DM_{in})		**DAOF**	**Blank**
Total		11.2 ± 2.9	13.1 ± 1.7

[a] Components that are present in the DM after drying.

[b] Corrected to account for the presence of non-ash DM originating from the reducing agent.

[c] Acid-insoluble lignin.

[d] Calculated assuming that lignin oil, besides phenolic monomers, only comprises oligomers.
[e] Components that are not present in the DM after drying.

Table S7 Phenolic monomer composition of the lignin oil isolated from the organic phase obtained after treating MWL (0.63 g), with and without the addition of formic acid (0.11 g), in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature), in the presence of $Na_2S_2O_4$ (1 g), for a duration of 3 hours.

Component	MWL Yield (wt%)[a]	MWL + formic acid Yield (wt%)[a]
Syringol	0.2	0.2
4-Propenyl guaiacol	0.7	0.5
Acetoguaiacone	-	0.1
Guaiacylacetone	0.2	0.1
Guaiacylpropan-1-one	-	-
4-Propyl syringol	-	-
Syringaldehyde	0.4	0.5
4-Propenyl syringol	3.4	3.0
Acetosyringone	1.4	1.6
Desaspidinol	0.1	0.2
4-Ethoxy-3,5-dimethoxybenzaldehyde	0.9	0.7
Sinapyl alcohol	0.1	0.0
Total	7.6	6.8

[a] Expressed with respect to the weight of MWL introduced in the reactor.

Table S8 Recovery of sulfur in the solid, organic and aqueous fractions obtained from the treatment of birch sawdust (3 g, particle size < 2 mm) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature),) for a duration of 3 hours, in the presence of 1 g of $Na_2S_2O_4$ (DAOF). The reactions were carried out in triplicates. The analysis of the sulfur content in each fraction was performed via ICP-AES.

Fraction	Recovery (wt%)[a]
Solid	4.0 ± 0.3
Organic	50.5 ± 2.5
Aqueous	27.4 ± 0.8
Total	81.9 ± 3.6

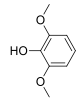
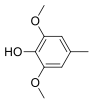
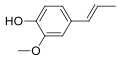
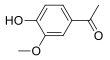
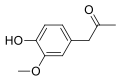
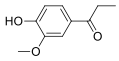
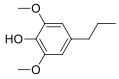
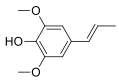
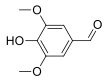
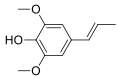
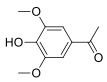
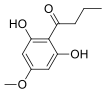
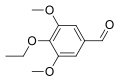
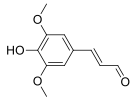
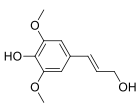
[a] Expressed with respect to the amount of sulfur contained in 1 g of $Na_2S_2O_4$

Table S9 Recovery of the butanol co-solvent in the organic and the aqueous fractions obtained from the treatment of birch sawdust (3 g, particle size < 2 mm) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature),) for a duration of 3 hours, in the presence of 1 g of $Na_2S_2O_4$ (DAOF), or in the absence of $Na_2S_2O_4$ (Blank). The reactions were carried out in triplicates.

Fraction	DAOF Recovery (wt%)[a]	Blank Recovery (wt%)[a]
Organic	90.6 ± 0.2	88.9 ± 0.7
Aqueous	6.7 ± 0.0	7.2 ± 0.3
Total	97.3 ± 0.3	96.1 ± 1.0

[a] Expressed with respect to the weight of butanol introduced (for the reaction and subsequent washing).

Table S10 GC-MS identification of the peaks for phenolic monomers detected in the lignin oil isolated from the organic and the aqueous fractions obtained for the dithionite-assisted organosolv fractionation (DAOF) and for the organosolv treatment of birch sawdust (3 g, particle size < 2 mm) in a mixture of n-butanol and water (120 mL, 50% v/v), at 200 °C, under 30 bar of N_2 (introduced at ambient temperature), for a duration of 3 hours. Unless otherwise reported, peak identification was based on mass spectral database searches using the NIST MS Search 2.2 software.

Component	Structure	Weighed match (%)	Reverse match (%)
Guaiacol[a]		91	91
Syringol[a]		90	90
Methyl syringol[a]		86	89
4-Propenyl guaiacol[a]		93	93
Acetoguaiacone[a]		90	90
Guaiacylacetone[a]		88	89
Guaiacylpropan-1-one		87	88
4-Propyl syringol[b]		-	-
4-Propenyl syringol (I)		90	90
Syringaldehyde[a]		83	83
4-Propenyl syringol (II)		94	94
Acetosyringone[a]		93	93
Desaspidinol		82	83
4-Ethoxy-3,5-dimethoxybenzaldehyde		80	81
Sinapyl aldehyde		91	91
Sinapyl alcohol		92	93

[a] Peak identification was further confirmed by comparison with pure standards.

[b] Peak identification was based on spectra reported elsewhere.[17]

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