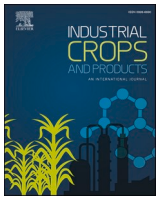
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OrganoCat fractionation of vine shoots for coproduction of bioethanol, furfural, and lignin

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ABSTRACT

Treatment of vine shoots with the OrganoCat method, using a mixture of oxalic acid (0.1 M) in water and 2-methyltetrahydrofuran (1:1) at 140 °C for three hours, allowed a separation of the lignocellulosic biomass into three fractions: an undissolved pulp, an aqueous solution and a solid obtained by evaporation of the organic phase. The resulting pulp was enriched in cellulose with greater crystallinity than the initial one due to the dissolution of the amorphous part, which facilitated enzymatic digestion and produced a significantly high glucose content, close to 70 g/L, and an excellent substrate for ethanol production, reaching levels of 35 g/L. Lignin, was recovered from the organic phase, and hemicellulose-derived sugars in the aqueous phase were converted into furfural (64 % yield from xylose) and ethanol (98 % yield from glucose). Overall, we demonstrate that the OrganoCat method allows an interesting fractionation of the shoots and also enables the full utilization of the obtained fractions that can be efficiently transformed into high-value renewable products such as bioethanol, furfural and high-purity lignin, highlighting the potential of the method as a promising pretreatment technique for the valorization of lignocellulosic biomass.

1. Introduction

As the demand for energy and chemicals continues to rise, the search for renewable and sustainable sources has become a priority. Currently, there is no technology capable of completely replacing fossil fuels, so it is necessary to find resources that can gradually reduce our dependence on them (Holechek et al., 2022; Wang et al., 2021; Wang and Azam, 2024). The use of biomass presents a promising solution because it is abundant and renewable and offers several environmental and economic advantages.

The biorefinery concept involves the use of biomass as a raw material to produce a range of products and energy in a sustainable manner. One of the most commonly utilized forms of biomass is lignocellulosic biomass (LB), which is rich in carbon and is mainly composed of three polymers that are linked by an ordered matrix, i.e., cellulose, hemicellulose and lignin in different proportions depending on the type of lignocellulosic biomass used (Kassim et al., 2020; Patel and Shah, 2021). Cellulose is a complex carbohydrate and the main structural component of plant cell walls. It is a polysaccharide made up of glucose repeat units linked together by β-1–4-glycosidic bonds. Hemicellulose, which is the second most abundant polysaccharide after cellulose (15 – 30 % by weight of the lignocellulosic biomass) (Kapu and Trajano, 2014), is a short and branched polymer with units of different C5 and C6 polysaccharides. It is linked to cellulose by hydrogen bonds and to lignin by covalent bonds; this allows it to be easily transformed into other products, especially after heat treatment (Yogalakshmi et al., 2022). Lignin is composed of an intricate network of phenylpropane units, primarily coniferyl, sinapyl, and p-coumaryl alcohol, which provides rigidity and strength to plant cell walls and the structural support for plants (Fernández-Rodríguez et al., 2017).

Among the various LB resources, the wastes from widely planted agricultural crops are promising sources of abundant materials for bio-based production. Vineyards are good examples of this, and their global cultivation area, including all vine plantations used worldwide for various purposes, was estimated to be approximately 7.2 million hectares in 2022 (OIV, 2023). This substantial vineyard area generates a

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significant amount of vine shoot (VS) residue, which may be utilized in an alternative manner. Currently, it is often left in vineyards or burned, leading to significant environmental issues (Cebrián-Tarancón et al., 2021; Contreras et al., 2022).

LB residues such as VS could be processed to generate bioproducts and biofuels, such as e.g bioethanol, provided that an adequate transformation process is defined. Bioethanol accounts for 53 % of the global biofuel used in transport biofuels and is a key element ensuring national energy and economic security (IEA, 2023). Ethanol from LB (cellulosic ethanol) has received global attention as a strategic alternative fuel to gasoline, produced from LB feedstocks that do not compete with food crops, which has driven intense R&D in production technology that has resulted in the operation of pre-commercial facilities (Raj et al., 2022). The process of cellulosic ethanol production comprises i) raw biomass pretreatment, ii) enzymatic hydrolysis of the cellulose rich-pretreated material and, iii) fermentation of the monomeric sugars released from the carbohydrates into ethanol during the hydrolysis step. Despite the significant advances achieved in the development of cellulosic ethanol technology, there are still technical challenges for different process steps (Padella et al., 2019). One of them is related to fractionation of the raw LB that should be able to generate the different biomass components (mostly cellulose, hemicellulose, and lignin) separately for valorization, thus approaching a real biorefinery concept. In this context, the development and application of new pretreatment technologies continues to arouse great interest within the scientific community (Zhou and Tian, 2022). There are different strategies for the fractionation and obtaining of the main LB components (Basak et al., 2023; Djellabi et al., 2023; Hu et al., 2023; Mankar et al., 2021; Prasad et al., 2023; Roy and Chundawat, 2023; Sai Bharadwaj et al., 2023; Wang et al., 2025; Zhou et al., 2023), however while other studies demonstrate effective strategies for biomass fractionation, many of them are carried out under harsh conditions of pressure and temperature, resulting in degradation of LB and formation of reaction byproducts. The method proposed in this work, called OrganoCat (Yan et al., 2014), offers notable distinctions, OrganoCat may provide improved selectivity in component separation, potentially leading to higher purity cellulose, hemicellulose, and lignin fractions and could operate under milder reaction conditions or utilize a more benign solvent system, potentially reducing energy costs and environmental impact. Moreover, the inherent catalyst reusability in the OrganoCat method presents an economic and sustainability advantage over systems that require solvent recovery or replacement, as highlighted in both studies. Thus, OrganoCat represents a promising alternative for efficient and sustainable biomass fractionation and the valorization of its components.

The OrganoCat method uses biphasic solvents with an acid catalyst (Cai et al., 2024; Gao et al., 2023; Xie et al., 2025). It was based on the depolymerization of hemicellulose to form an aqueous stream of structural carbohydrates, where the lignin is extracted into the organic phase and the cellulose remains as a suspended solid, which would be used to obtain fermentable sugars for ethanol fermentation due to at moderate temperatures (80 – 140 °C), oxalic acid effectively catalyzes the breakdown of hemicellulose into soluble sugars in an aqueous solution, whereas the more crystalline cellulose pulp remains solid and unaffected by the acid catalysis (Grande et al., 2024; Langletz et al., 2024; Martinez Diaz et al., 2024; Schoofs et al., 2024). Remarkably, the concept of this method would allow both the catalyst and the solvent to be recovered and reused after the reaction. The organic solvent 2-methyltetrahydrofuran (2-MeTHF) is usually used, and organic acids such as oxalic acid or 2,5-furandicarboxylic acid (FDCA) are used as catalysts, with the reaction conditions varying between 120 – 160 °C for 1 – 6 h, where the best results of lignin separation were at 140 °C during 3 h (Grande et al., 2015; Schoofs et al., 2021). These catalysts accelerate the cleavage of hemicellulose bonds, releasing pentose sugars into the aqueous solution. This is particularly relevant since valorization of the hemicellulose fraction plays a crucial role in biorefinery processes based on lignocellulosic feedstocks to obtain biofuels and renewable chemicals. One example of bio-based chemicals is furfural, which is one of the most valuable compounds obtained from biomass in general and lignocellulosic materials in particular (Bozell and Petersen, 2010). Furfural is involved in a wide range of applications: solvents, pharmaceutical products, and biofuels (2-methylfuran and 2-methyltetrahydrofuran) (Mariscal et al., 2016). Currently, furfural is obtained from the pentoses of biomass through a process consisting basically of acid catalysis (for hydrolysis of hemicelluloses) and dehydration to furfural (Mariscal et al., 2016). Finally, OrganoCat method allows the recovery of a lignin-rich solid that could be used to obtain compounds with applications in different industrial sectors.

Previous works have reported different technologies to fractionate vine shoots and valorize their main components. Cardoza et al., (2004) fractionated vine shoots by a sequential acid/organosolv pretreatment to produce a high-purity organosolv lignin and bioethanol from C5 and C6 sugars and Castro et al. (2023) reported the valorization of steam-exploded vine shoots by the biological conversion of cellulosic glucose into hydrogen and the chemical conversion of hemicellulosic sugars into furfural (Cardoza et al., 2024; Castro et al., 2023). In this work, an integrated approach for the holistic use of relatively unexplored LB source VS for the production of several targeted bio-based products is studied. The pretreatment method used, the OrganoCat process, will yield three fractions; i) a cellulosic-rich solid, which is transformed into bioethanol through enzymatic hydrolysis and anaerobic fermentation, ii) a hemicellulosic liquor that is used for bioethanol and furfural production and, iii) a lignin-rich solid fraction of high purity that can be further valorized into different compounds. The various process steps, i.e., OrganoCat pretreatment at optimized reaction conditions (Grande et al., 2015), enzymatic hydrolysis of the cellulose-rich solid using commercial enzymes, ethanol fermentation of sugars released by enzymatic hydrolysis (EH) with *Saccharomyces cerevisiae* and furfural production under microwave with $AlCl_3$ as catalyst are tested, and the corresponding parameters to assess process efficiency calculated.

2. Materials and methods

2.1. Materials and chemicals

All reagents were used without further purification. The solvents 2-methyltetrahydrofuran (2-MeTHF) and oxalic acid were purchased from Merck-Sigma Aldrich. Acetone and ethanol were acquired from Scharlab. The standards used in FTIR characterization were Cellulose Avicel PH-101 from Fluka Analytical and Xylan from Beechwood (Hemicellulose), and Lignin (alkaline) from TCI.

2.2. Analysis of lignocellulosic biomass

The methods used to characterize the vine shoots were adapted from the standard method published by the National Renewable Energy Laboratory (NREL; Colorado, USA) (Sluiter et al., 2004). The main components of the biomass, i.e., cellulose, hemicellulose, and acid-insoluble lignin, were analyzed, together with minor components, extractives, acid-soluble lignin, acetyl groups, and ash. Briefly, the samples are first submitted to an extraction step, both in water and ethanol, to generate an extractives-free sample that is used in the following procedures. For carbohydrates and lignin content quantification, the samples underwent two acid hydrolysis steps, first with 72 % H_2SO_4 at 30 °C for 60 min and then, the samples were diluted with water to 4 % H_2SO_4 and put in an autoclave at 121 °C for 60 min. After cooling and filtration, the liquid is analyzed for sugars and acetic acid using High Performance Liquid Chromatography (HPLC), as described elsewhere Padilla-Rascón et al., (2021). (Padilla-Rascón et al., 2021) Furthermore, acid-soluble lignin was determined by UV-spectrophotometric analysis, and acid-insoluble lignin in the non-hydrolyzed biomass was determined by weight, according to the specific protocols. Ash content was

measured by calcination the biomass sample at 575 °C for 6–8 h. Table 1 summarizes the average values for the dried samples. The components in the lignocellulosic biomass were analyzed in triplicate.

2.3. Treatment of lignocellulosic biomass

The Organocat reaction (Fig. 1) was carried out in batches in a mechanically stirred 300 mL thermostatic Teflon-lined steel Berghof reactor. In a typical run, 10 g of the lignocellulosic biomass and the corresponding quantity of catalyst were used to obtain a concentration of 0.1 M by suspending the mixture in 100 mL of water (Schoofs et al., 2021). Later, 100 mL of the organic solvent 2-MeTHF was added. The reactor was heated to 140 °C, stirred, and the reaction began. After three hours, the reactor was cooled on ice. Firstly, the pulp (Solid I) was isolated by filtration and the two liquid phases were reserved. The Solid I was washed with approximately 200 mL of water (4 ×50 mL) until the mother liquor obtained was completely transparent. Subsequently, it was washed with approximately 200 mL of acetone (4 ×50 mL), this caused the mother liquor still to be colored because there was lignin present in the solid, and the washing was repeated until the last mother liquor was transparent. The solvent of the washing procedure was evaporated at reduced pressure in a rotary evaporator, and a new solid (Solid III) was obtained.

Fifty milliliters of $CaCl_2$ (0.1 M) was added to the liquid phases to eliminate the acid catalyst. The addition of $CaCl_2$ allows the separation of the catalyst from the solution due to the formation of calcium oxalate. The solid was separated by filtration in a funnel. The aqueous liquids were analyzed by HPLC. The chromatographic separations were carried out in a Hi-PLEX H column at 60 °C using 0.6 mL/min sulfuric acid aqueous solution (0.01 M). The solvent of the organic liquid phase (2-MeTHF) was evaporated under reduced pressure in a rotary evaporator, the obtained solid is called Solid II.

All solids were collected, dried at room temperature, and characterized by X-ray diffraction (XRD) and Fourier transform infrared spectroscopy (FTIR).

Solid I (cellulose- rich pulp fraction) was obtained after filtering and washing the filtration solid left after the OrganoCat treatment with distilled water. Solid II (lignin) was obtained after evaporation of the organic solvent. Finally, a third solid, Solid III, was obtained after cleaning the pulp (Solid I) with acetone, recovering the mother liquor and evaporating it in a rotary evaporator. The characterization data demonstrated that Solid III was also lignin.

2.4. Enzymatic hydrolysis tests

The pulp or Solid I obtained after the OrganoCat treatment was dried at room temperature and used as substrate in enzymatic hydrolysis (EH). The EH tests were carried out in 100 mL Erlenmeyer flasks containing different substrate concentrations (5 – 15 and 20 % w/w) at pH 5 (0.05 M sodium citrate buffer). The flasks were incubated in an orbital shaker at 50 °C for 72 h. Cellic® CTec2 (Protein content was 200 mg/mL (determined by BCMTM Protein Assay Kit- Reducing Agent Compatible. Ref 23250, Sigma Aldrich) was used as an enzyme cocktail with various enzyme loadings (15, 30 and 45 FPU/g cellulose). Tween 80 (25 mg/g substrate) was added to prevent irreversible adsorption of the enzyme to the pretreated substrate, thus improving the enzymatic accessibility. Samples were taken periodically, and glucose and xylose contents analyzed via high-performance liquid chromatography (HPLC) with a chromatograph equipped with a refractive index detector and a carbohydrate column (CARBOSep CHO-782 Pb); ultrapure water was used as the eluent with a flow rate of 0.6 mL/min and a column temperature of 70 °C.

The EH yield was expressed as a percentage of the maximum theoretical yield and calculated as follows:

$$\text{EH yield}(\%) = \frac{\text{g glucose by EH}}{\text{g glucan in pulp} \times 1.11} \times 100$$

Table 1
Composition (wt %) of vine shoot biomass dried at 105 °C.

Compound	Vine shoots
Extractives	8.3 ± 0.2
Aqueous	7.1 ± 0.4
Organic	1.2 ± 0.25
Cellulose	32.5 ± 0.5
Hemicellulose	18.4 ± 0.3
Hemicellulose sugars (monomeric sugars)	
Xylose	16.7 ± 0.3
Galactose	2.1 ± 0.07
Arabinose	1.1 ± 0.04
Mannose	0.80 ± 0.01
Insoluble Lignin	25.4 ± 0.1
Soluble Lignin	1.6 ± 0.05
Ash	3.6 ± 0.3
Acetyl Groups	5.3 ± 0.05

2.5. Ethanol production from the OrganoCat pretreated material

2.5.1. Ethanol production from the pulp

Romero-Garcia et al. (2022) described the presaccharification and simultaneous saccharification and fermentation (PSSF) process used for ethanol production from Solid I obtained after OrganoCat treatment with *Saccharomyces cerevisiae* "Ethanol Red" (Fermentis, France). Fed-batch mode was used with a 15 % (w/w) initial solid concentration, and 5 % (w/w) additional substrate was added after 24 h. Samples were taken periodically and analyzed for ethanol and glucose as described in Moreno et al., (2021). (Moreno et al., 2021; Romero-García et al., 2022)

2.5.2. Ethanol production from the aqueous phase

S. cerevisiae described above was also used to ferment the glucose present in the aqueous phase. Yeast inoculum was prepared in synthetic glucose media consisting of (g/L): yeast extract, 5; NH_4Cl, 2; KH_2PO_4, 1; $MgSO_4{\cdot}7\ H_2O$, 0.3; and glucose, 30, as described by López-Linares. (López-Linares et al., 2014)

The fermentation tests were carried out in triplicate at 35 °C in 100 mL glass flasks with working volumes of 25 mL, and an orbital shaker was operated at 150 rpm for 24 h. The pH of the liquor was adjusted to pH 5 with NaOH prior to fermentation. A yeast inoculum of 4 % (v/v) was added, corresponding to an initial cell concentration of 0.25 g/L, which was determined with the absorbance at 620 nm. (Padilla-Rascón et al., 2021) At the end of the fermentation, the broth was centrifuged at 11,500 rpm for 10 min and analyzed by HPLC to determine the ethanol and sugar contents, as indicated in Section 2.5.1. The ethanol yield was determined as g ethanol/g consumed glucose and as a percentage of the theoretical ethanol yield (0.51 g ethanol/g glucose).

After fermentation, the ethanol was recovered from the broth by vacuum distillation at low temperature (40 °C), and then, the evaporated water was returned to maintain the initial xylose concentration in the liquor (xylose-rich liquor).

2.6. Microwave treatment for furfural production

The furfural production experiments were performed in triplicate with an Anton Paar microwave reactor (Monowave 400), as described elsewhere. (Padilla-Rascón et al., 2021) The liquor was filtered through a 0.45 µm membrane. Volumes of 4 mL were placed in 10 mL glass tubes with magnetic stirring at 600 rpm. $AlCl_3$, a Lewis acid, and 2 % w/v H_2SO_4, a Brønsted acid, were used as catalysts. Chemical conversion of the xylose in the liquor was performed at different temperatures (170 and 200 °C) and with various $AlCl_3$ concentrations (0.1, 0.2, and 0.3 M). The reaction was also carried out without any catalyst at both temperatures assayed. In each test, the liquid was heated to the desired temperature and immediately cooled to room temperature with compressed

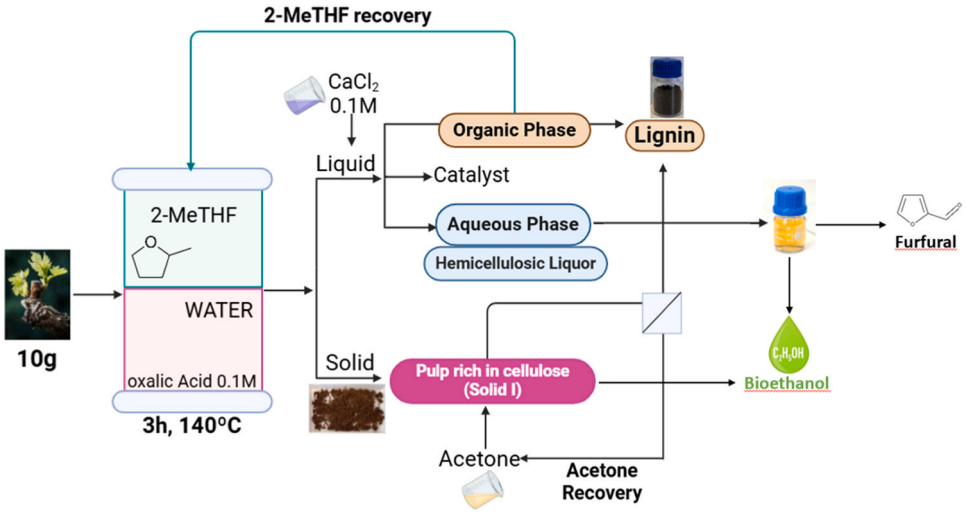


Fig. 1. Schematic for the OrganoCat treatment.

air. The concentrations of residual xylose, furfural and formic acid in the final liquids were measured by HPLC. The xylose conversion, furfural yield and furfural selectivity were calculated as follows (Padilla-Rascón et al., 2021):

$$\text{Xylose conversion } (\%) = \frac{\text{moles of xylose consumed}}{\text{moles of initial xylose}} \text{ x } 100$$

$$\text{Furfural yield}(\%) = \frac{\text{moles of furfural produced}}{\text{moles of initial xylose}} \text{ x } 100$$

$$\text{Furfural selectivity } (\%) = \frac{\text{moles of furfural produced}}{\text{moles of xylose consumed}} \, x\, 100$$

2.7. Biomass characterization

X-ray diffraction (XRD). Diffractograms of pulp, lignin and original biomass were obtained with an X'Pert Pro PANalitical diffractometer equipped with a CuKα radiation source (λ = 0.15418 nm), and an X'Celerator detector using real-time multiple-strip (RTMS) detection with real-time multiple strip (RTMS) technology was used to record the X-ray diffraction profiles of the samples. The diffraction patterns were recorded in steps over a range of Bragg angles (2θ) between 4° and 90° at a scanning rate of 0.02° per step and an accumulation time of 50 s after grinding and placing the samples on a stainless-steel plate. The diffractograms were analyzed with X'Pert HighScore Plus software.

Fourier-Transform Infrared (FTIR) spectroscopy. A Jasco FT/IR-6300 spectrophotometer was used with pellets of KBr mixed with the different samples to obtain the infrared spectra. A total of 180 scans were performed in transmittance mode with a resolution of 4 cm^{-1} over the frequency range 4000 – 450 cm^{-1}.

Scanning electron microscopy (SEM). SEM micrographs of different vine shoots (untreated and treated) samples were analyzed with a Hitachi S-3000 N instrument. The samples were treated with increasing concentrations of ethanol:water (10:90, 50:50, 75:25 and 100:0) to fix the structure and dehydrate the samples. Then, they were metallized in a Balzers SCN 004 gold sputter coater with a thin layer of gold.

3. Results and discussion

3.1. OrganoCat treatment method

The Organocat method is used to separate the components of lignocellulosic biomass. After 3 h of reaction at 140 °C, using oxalic acid as a catalyst and Water:2-MeTHF 1:1, we obtain in the aqueous phase, the hemicellulose depolymerized into sugar monomers. This acid is capable of hydrolyzing the hemicellulose and depolymerizing it, but it is not strong enough to depolymerize the cellulose. In the organic phase, the lignin is dissolved and recovered as a solid (Solid II) after evaporation of 2-MeTHF and as pulp a solid is obtained (Solid I), which after washing it with acetone, is cleaned of lignin that has not been carried over to the organic phase (Solid III) (Grande et al., 2024, 2015; Martinez Diaz et al., 2024; Schoofs et al., 2024, 2021).

Firstly, to evaluate the fractional capacity of the OrganoCat treatment, the mass of each recovered solid was calculated to compare it with the theoretical total for cellulose, hemicellulose and lignin (initial biomass), and the expected amounts were calculated for Solid I with cellulose and Solid II and Solid III with lignin.

The total biomass percentage recovered after the OrganoCat treatment was close to 65.9 % based on the starting biomass, partitioned into 55.4 % cellulose-rich pulp (Solid I) and 10.5 lignin (Solid II and Solid III), considering that hemicellulose dissolved in the aqueous phase. Therefore, we characterized each of the solids obtained to investigate the structure and composition of each fraction.

3.1.1. Characterization of solids obtained with the OrganoCat method

The solids obtained for the OrganoCat treatment were characterized by FTIR, XRD, and SEM. The results are described in the characterization section of the Supplementary Information. Spectroscopy was used in this work to study the functional groups of the cellulose, hemicellulose, and lignin obtained during the treatment. The spectra of Solid I, Solid II, and Solid III are shown in Figure S1 and compared with those of commercial standards between 1800 and 800 cm^{-1}, the fingerprint region for lignocellulosic biomass. Figure S1A shows the FTIR spectrum for Solid I compared to the cellulose, hemicellulose and lignin standards, and Figure S1B shows the FTIR spectra for solids II and III along with the lignin pattern.

The presence of a peak at 1508 cm^{-1} is usually taken as an indication of the presence of lignin, especially when a weak band at 1520 cm^{-1} is present. This was observed for Solid II and Solid III as well as the pulp (Solid I), which confirmed the material balance (Table 2 and Table 3) and indicated that there were traces of lignin in the pulp. In addition, typical cellulose and hemicellulose peaks were observed, such as those at 1336 cm^{-1} (C-H vibrations in cellulose), 1157 cm^{-1} (C-O-C vibrations in cellulose and hemicellulose), 1048 cm^{-1} (C-O stretching in cellulose and hemicellulose), and 897 cm^{-1} (C-H deformation in cellulose); these peaks were characteristic of polysaccharides. (Hergert, 1960; Pandey and Pitman, 2003; Sun et al., 2001, 2005).

In addition to the peak at 1508 cm^{-1}, other lignin peaks were observed, such as those at 1035 cm^{-1} and 1085 cm^{-1} peaks (C-O deformations in aliphatic ethers and guaiacyl aromatic methoxyl groups, respectively); 1118 cm^{-1} (aromatic skeletal vibrations); 1264 cm^{-1}

Table 2
Composition of the pulp (Solid I) calculated with the NREL standard methods and HPLC results for the aqueous phase.

Compound	Pulp (% dry weight)	Aqueous phase (100 mL)	
Cellulose	44.6 ± 0.8	Xylose (g/L)	11.1 ± 0.1
Hemicellulose	6.4 ± 0.1	Glucose (g/L)	7.54 ± 0.13
Hemicellulosic sugars		Galactose (g/L)	1.99 ± 0.06
Xylose	6.7 ± 0.1	Arabinose (g/L)	1.27 ± 0.03
Galactose	0.2 ± 0.03	Mannose (g/L)	0.79 ± 0.06
Arabinose	< 0.1		
Mannose	0.5 ± 0.01	Acetic Acid (g/L)	1.11 ± 0.02
Insoluble Lignin	35.7 ± 0.3	Formic Acid (g/L)	0.39 ± 0.05
Soluble Lignin	1.5 ± 0.05		
Ash	4.6 ± 0.1		
Acetyl Groups	1.0 ± 0.03		

Table 3
Mass Balance of the Organocat method.

	Initial sample	Solid I	Solid II and III*	Aqueous phase	% BM
Sample weight (g)	10.00	5.54	1.05	100 mL	-
Glucose from Cellulose (g)	3.61	2.74	0.00	0.75	96.7 %
Sugars from Hemicellulose (g)	2.07	0.42	0.00	1.52	93.7 %
Lignin (g)	2.54	1.98	1.05	0.00	119.3 %

The amount of Solid III obtained is 20 % of the total

(guaiacyl ring breathing, C-O stretching in lignin and C-O linkages in guaiacyl aromatic methoxyl groups); 1318 cm^{-1} (C-O vibration in syringyl derivatives); 1457 cm^{-1} (C-H deformation in lignin); 1593 cm^{-1} and 1508 cm^{-1} (C═C skeletal vibrations (aromatic ring) in lignin and 1658 cm^{-1} (absorbed O-H and conjugated C-O in lignin). The presence of the peak at 1508 cm^{-1} and the weak band at 1520 cm^{-1} was a clear indication of the presence of lignin (Hergert, 1960; Pandey and Pitman, 2003; Sun et al., 2001, 2005). In summary, the spectrum of Solid I had peaks for the three major components, so complete separation was not carried out. Solids II and III are quite similar to the Kraft lignin pattern used, which allows us to confirm that it is lignin.

The X-ray diffraction profile for the pulp (Solid I) was compared with the original biomass profile, as shown in Figure S2. The profiles of both samples were very similar except for significant peaks in the pulp sample that were due to possible catalyst residues. The prominent peak at 23 ° that corresponded to the (200) reflection planes and a band between 15 – 17 ° for the combined (110) and (1−10) reflections were observed for both samples, so it was deduced that there was no change in the biomass structure after treatment. (French, 2014; French and Santiago Cintrón, 2013; Morales-delaRosa et al., 2014) The crystallinity index (CI) was measured with the following expression after normalizing the XRD profiles:

$$CI = \frac{(I_{23} - I_{18})}{I_{23}} \quad x \quad 100$$

where I_{23} and I_{18} are the intensities of the 23 ° and 18 ° peaks, respectively. The CI of the original vine shoots was relatively low at 51 % due to the presence of amorphous components in the lignocellulosic biomass. After the OrganoCat treatment, the CI of the solid obtained (pulp) reached 62 %. This increase was due to the dissolution of some of the amorphous components in the biomass, leaving the most crystalline fraction in the solid, which was more recalcitrant to treatment, reaction, or dissolution. The morphologies of the untreated vine shoots and the pulp obtained after the OrganoCat treatment (Solid I) were examined with SEM (Fig. 2). When the samples were compared by SEM, no significant morphological changes were observed. The characteristic vascular structures of the original biomass remained intact, with only the presence of surface fibers noticeable in the solid material. The SEM micrographs confirmed the data obtained by XRD and the crystallinity measurements that the initial structures of the samples had not been modified, but some of the lignin and almost all of the hemicellulose were dissolved. These results differed from those observed in other treatment methods of lignocellulosic biomasses, in which the fragmented cellulose exhibited lower crystallinity and loss of the original structure. (Lara-Serrano et al., 2023, 2020, 2019; Morales-delaRosa et al., 2014)

An X-ray powder diffraction analysis was also carried out to study the morphology of the lignins obtained and it was compared with the original biomass. The diffractograms (Figure S3) showed a large area

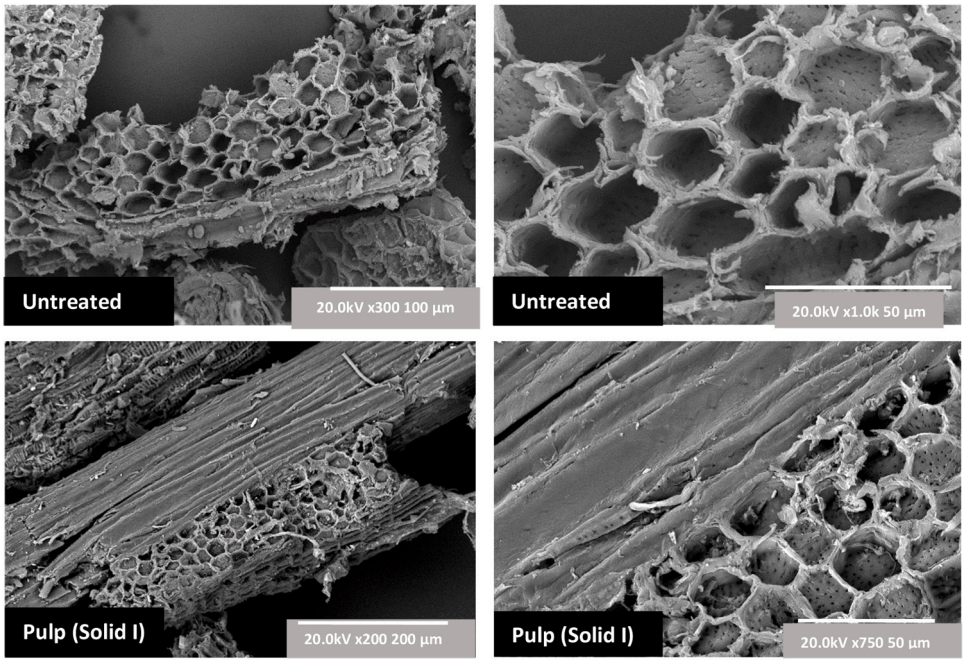


Fig. 2. SEM micrographs of the untreated vine shoots and pulp obtained after the OrganoCat treatment (Solid I). The left panels depict the samples at lower magnifications, and the right panels depict the samples at higher magnifications.

under the curve for lignins, confirming that both samples are amorphous and there are no remains of original biomass.

3.1.2. Composition of the pulp

The pulp composition (Solid I) was determined with the NREL standard methods, (Sluiter et al., 2010) and the aqueous phase was analyzed by HPLC (Table 2). The results confirmed that full fractionation was not achieved because there was still lignin and hemicellulose in the sample. Solid I was enriched in cellulose with respect to the starting material (vine shoots, Table 1). The main compounds in Solid 1 were 45 % cellulose and 36 % lignin that was not dissolved (Table 2).

Based on this analysis, we performed a mass balance of the three main components in the vine shoot sample: cellulose, hemicellulose, and lignin. The recovery rates were quite high for all three components (Table 3). However, the amounts of glucose and hemicellulose sugars recovered were lower, and the weight of the lignin fraction was higher. These results indicated that small amounts of sugars reacted during the treatment to form humins, which remained in Solid I and were detected as lignin. Approximately 70 % of the initial cellulose was present in the pulp (Solid I) (in Table 3 shown as glucose), while 21.1 % was present in the aqueous phase as glucose. A total of 93.7 % of the hemicellulose was recovered, and 73 % was present as sugars in the aqueous phase, with 17 % remaining in the pulp (in Table 3, shown as hemicellulose-derived sugars). Finally, 41.3 % of the lignin was dissolved and recovered (Solids II and III), where the remaining lignin was in the pulp.

The aqueous phase was concentrated by vacuum distillation to obtain a sugar-rich liquid. The composition of the concentrated sample is shown in Section 3.4.

3.2. Enzymatic hydrolysis of pulp (Solid I)

The PSSF process comprises an enzymatic hydrolysis (EH) step under optimal enzyme conditions, followed by fermentation of the sugars released in the same reaction vessel. Thus, before ethanol production from the OrganoCat pretreated solid by PSSF, a study of the best EH conditions for maximizing sugar production was carried out. EH tests at relatively low substrate consistencies were carried out to evaluate the effects of different enzyme loadings on the enzymatic digestibility of Solid I. The glucose contents in the EH media and the EH yields obtained for different times (24, 48, and 72 h) and enzyme loadings (15, 30, and 45 FPU/g cellulose) with 5 % (w/w) pulp as the substrate are shown in Table 4.

The use of 15 FPU/g cellulose resulted in a low glucose concentration and EH yield (8.6 g/L and 34 % of the theoretical value, respectively). When the highest enzyme loading was tested, the glucose level increased up to 14.6 g/L (corresponding to approximately 57 % of the theoretical yield). A slightly lower concentration (13.1 g/L) was observed at an enzyme loading of 30 FPU/g cellulose. In comparison, Dávila et al. (2017) achieved 49 % total glucan conversion for enzymatic hydrolysis of autohydrolyzed vine shoots at 201 °C after 96 h, using an enzyme concentration of 25 FPU/g solid (cellulase and β-glucosidase, β-glucosidase/cellulase ratio of 5 IU/FPU) (Dávila et al., 2017). The OrganoCat treatment used in the present work provided advantages in terms of the enzymatic digestibility of the pretreated vine shoot biomass, which resulted in slightly more efficient carbohydrate conversions. On the other hand, the maximum concentrations reached for xylose were 1.5 – 2 g/L (data not shown), which corresponded to a xylan conversion of approximately 50 – 62 %.

To assess EH performance with high solid loadings, higher pulp concentrations (15 and 20 % w/w) were used in the EH tests with two different cellulase loadings (30 and 45 FPU/g cellulose). The results are shown in Fig. 3.

The maximum glucose concentration was reached at 72 h. When a 15 % (w/w) substrate consistency was used, 48 and 57 g/L of glucose were produced with 30 and 45 FPU, respectively (corresponding to 56 and 64 % of the theoretical yield). For the 20 % (w/w) substrate, the highest glucose concentrations were approximately 56 and 68 g/L for 30 and 45 FPU/g cellulose, respectively (corresponding to 45 and 54 % of the theoretical yield). It should be noted that when the enzyme loading was 30 FPU/g cellulose, only a slight increase in the glucose yield from 48 to 72 h (in both cases with 15 and 20 % substrate consistency) was produced.

The maximum xylose concentrations were 6.1 and 7.6 g/L when using 15 % substrate for 30 and 45 FPU/g cellulose, respectively. In the case of a 20 % substrate consistency, the xylose concentrations were 7.8 and 8.1 g/L for 30 and 45 FPU/g cellulose, respectively (data not shown). The xylan conversion yields ranged from 37 % to 47 % of the theoretical yield.

On the other hand, it should be noted that despite the relatively high glucose concentrations and EH yields achieved in comparison with similar reported data from Dávila et al., (2017), the fluidity of the enzymatic hydrolysate was not sufficient to carry out a fermentation process (Dávila et al., 2017). It is well known that the use of high solid

Table 4
Glucose contents in EH media (g/L) during incubation and EH yields for 72 h tests with 5 % (w/w) solid concentrations and different enzyme loadings.

Enzyme loading (FPU/g cellulose)	Glucose (g/L)			EH yield (% of theoretical)
	24 h	48 h	72 h	
15	7.1	8.2	8.6	34
30	10.1	12.7	13.1	51
45	10.8	14.2	14.6	57

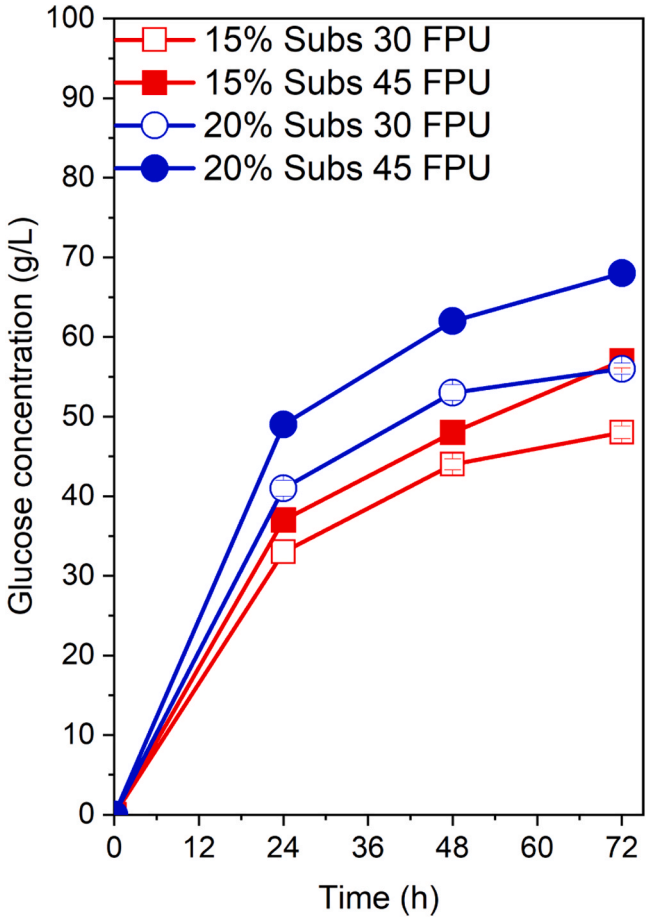


Fig. 3. Glucose concentrations reached in EH experiments at 15 % and 20 % (w/w) substrate consistency with two enzyme loadings (30 and 45 FPU/g cellulose). The results are the medians of three replicates. Error bars represent the standard deviations.

loadings during EH brings many challenges, such as increases in medium viscosity affecting the subsequent enzymatic hydrolysis yield (Koppram et al., 2014). However, it was found that the fed-batch process alleviated this and reduced the viscosity caused by the high solids. (Zhang et al., 2010; Zhao et al., 2013) Therefore, based on the previous results showing low final fluidity for the media in the 20 % (w/w) batch EH experiments, fed-batch EHs were used to improve the final fluidity of the enzymatic hydrolysate and favor subsequent fermentation. Fed-batch experiments were tested with 15 % (w/w) initial solid concentrations by adding 5 % (w/w) at 24 h and using 45 FPU/g cellulose as the enzyme loading. The results are shown in Fig. 4.

The highest glucose concentration (71 g/L) was achieved at 72 h after the onset of EH, with an approximate 56 % theoretical EH yield. However, it is important to note that at 48 h, more than 95 % of the maximum glucose concentration was reached (67 g/L). The glucose concentration was slightly higher than that observed when using 20 % substrate loading in batch mode, where approximately 68 g/L was attained (Fig. 3). In addition, a slightly higher glucose concentration and EH yield were observed in the fed-batch mode than in batch mode with a 20 % (w/w) substrate consistency, and a lower viscosity of the medium was also obtained. These results indicated that when using high substrate loadings, the fed-batch mode is better for obtaining a liquefied enzymatic hydrolysate for fermentation with different process configurations, such as separate hydrolysis and fermentation or simultaneous hydrolysis and fermentation.

3.3. Ethanol production from the pulp (Solid I)

The pulp was also employed for ethanol production. Based on the results observed in the EH section, a fed-batch PSSF process was used to produce ethanol from the solid phase and promote substrate liquefaction. PSSF was started with a 15 % (w/w) solid concentration, and an additional 5 % (w/w) was added at 24 h. After 48 h of presaccharification, the temperature was decreased to 35 °C, and the media were inoculated with *Saccharomyces cerevisiae* (1 g/L). The glucose concentrations at 24 and 48 h were 43 and 72 g/L, respectively. These results were consistent with those obtained previously in the fed-batch EH test, in which the maximum glucose concentration was approximately 70 g/L (Fig. 4). Regarding ethanol concentration, at 8 h of inoculation about 3.2 g/L were observed. Then, at 24 and 48 h after inoculation, the ethanol concentration increased to 6.8 and 33 g/L, respectively. Therefore, based on the ethanol concentrations obtained in the process, the ethanol yield reached approximately 70 % of the theoretical PSSF yield (Fig. 5).

The PSSF process has previously been used to produce ethanol from biomass pretreated substrates, and the results obtained herein support this. For example, several process configurations for ethanol production from olive stones subjected to sequential acid treatment followed by steam explosion have been tested (Romero-García et al., 2022), and PSSF was one of them. The assays were carried out at 15 and 20 % solids, and the results displayed ethanol concentrations and process yields similar to those obtained in the present work (35 g/L, and 65 % of theoretical) with 40 FPU/g of dry substrate. Likewise, a previous work

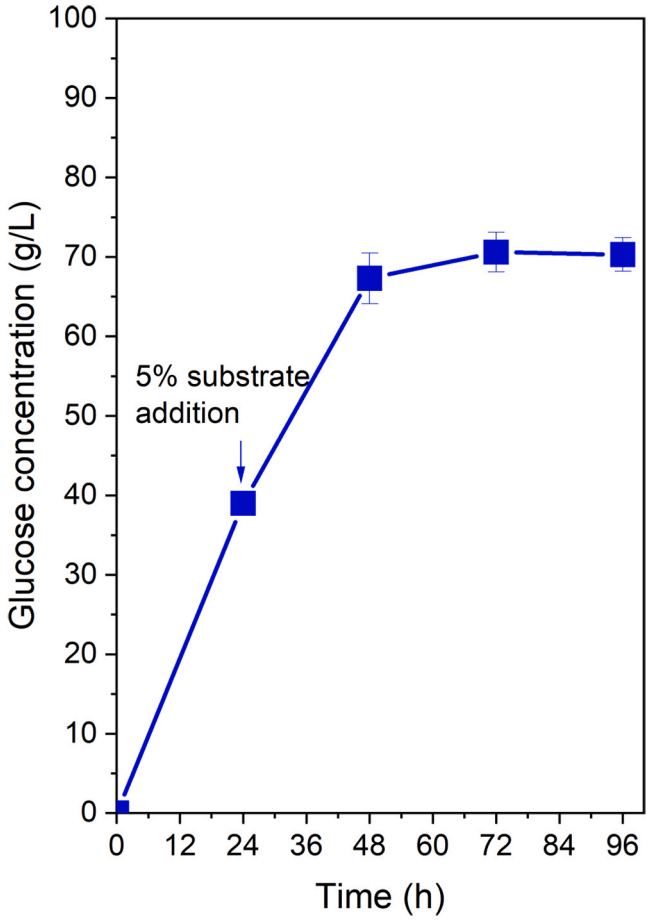


Fig. 4. Glucose concentrations reached in the fed batch EH test with 15 % initial solid concentration using 45 FPU/g cellulose and adding 5 % (w/w) solids at 24 h. The results are the medians of three replicates. The error bars represent standard deviations.

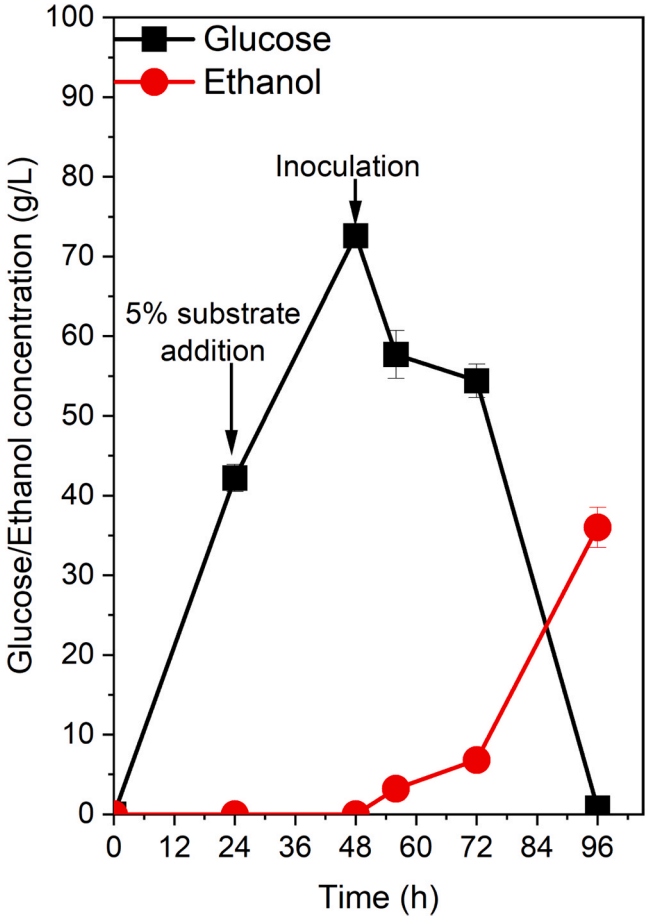


Fig. 5. Glucose and ethanol concentrations reached with the fed batch PSSF method at 15 % initial solid concentration using 45 FPU/g cellulose and adding 5 % (w/w) solids at 24 h. The results are medians of three replicates. The error bars represent standard deviations.

(Palacios et al., 2021) showed good performance from the PSSF process by producing ethanol from banana peel biomass previously pretreated in an autoclave at 121 °C for 15 min. The highest ethanol concentration (32.6 g/L) was reached for the PSSF process with a 25 % (w/v) solid load at 64 h with a commercial *S. cerevisiae* strain. Moreover, in the present work, the fed-batch mode was successful in using a high solid concentration of 20 % in the ethanol production process, which is otherwise particularly difficult for OrganoCat pretreated vine shoot biomass.

3.4. Valorization of the aqueous phase

The aqueous phase resulting from the OrganoCat treatment of the vine shoots was concentrated (hemicellulosic liquor) and used as a substrate to produce both ethanol and furfural. The compositions of sugars and inhibitors in this acid liquor (pH = 0.3) were as follows (g/L): glucose, 23.03 ± 0.08; xylose, 33.96 ± 0.11; galactose, 6.11 ± 0.03; arabinose, 3.89 ± 0.04; mannose, 2.43 ± 0.03; acetic acid, 3.42 ± 0.02; and formic acid, 1.20 ± 0.00. Considering its glucose and xylose content, the liquor was subjected to a first fermentation step with an ethanologenic yeast to use the glucose and leave a rich-xylose medium for furfural production via dehydration in a second step.

3.4.1. Ethanol production

Hemicellulosic liquors containing pentose and hexose sugars can be fermented by microorganisms able to assimilate both kinds of sugars. (Singh et al., 2025) In this work, *S. cerevisiae* yeast was selected because this step aims to convert only glucose, maintaining xylose in the medium to be chemically converted to furfural in the next process step. The hemicellulosic liquor was fermented by *S. cerevisiae* without prior detoxification after adjusting the pH to 5. The yeast assimilated almost all of the glucose at 24 h, and only 0.5 g/L remained in the fermentation broth after that time. Galactose in the liquor was partly assimilated by the yeast. The limited ability of *S. cerevisiae* to produce ethanol from galactose has been widely reported. (Park et al., 2014) An ethanol concentration of 11.3 g/L was obtained with the bioreactor, corresponding to an ethanol yield of 0.50 (98 % of the theoretical yield) and a productivity of 0.47 g/L/h. Using the same raw material, a similar ethanol yield was reported in the co-fermentation of the acid prehydrolysate with *E. coli* after detoxification with resins or NH_4OH. (Cardoza et al., 2024)

3.4.2. Chemical conversion of xylose into furfural

Microwaves have been revealed as an advantageous technology *versus* the conventional heating methods because microwave radiation produces a rapid and uniform heating due to ionic conduction, and this results in more efficient reactions. Different studies have reported better furfural yields using microwave-assisted reactions compared to traditional heating. (Padilla-Rascón et al., 2020)

In this work, the liquid obtained after glucose fermentation and ethanol distillation (xylose-rich liquor), which contained 31.19 ± 0.16 g/L xylose, was used for furfural production using a microwave reactor. The effects of the temperature and $AlCl_3$ concentration on the chemical conversion of xylose into furfural were studied. The potential of salts as catalysts in this conversion process, especially metal chlorides such as $FeCl_3$ or $AlCl_3$, has been widely studied. (Ershova et al., 2017) These salts act as Brønsted acids and favor the conversion of xylose into furfural by dehydration. (Schwiderski et al., 2014)

In general, the results indicated that higher treatment temperatures enhanced furfural production (Table 5). Thus, when the reaction was carried out at 170 °C, poor xylose conversion resulted, and the furfural yields were lower than 50 % even with the highest $AlCl_3$ concentration (run 4, 49.5 % yield). The furfural concentrations ranged from 4.1 to 9.9 g/L at 170 °C and from 9.4 to 12.8 g/L at 200 °C. The highest furfural production rate was achieved at 200 °C with 0.1 M $AlCl_3$, which corresponded to a yield of 64 % (run 6). However, similar results were obtained at that temperature without salt addition (run 5), which was advantageous even though 6.8 g/L residual xylose remained in the liquid in the latter case. Probably, the effect of the temperature could have compensated for the absence of the catalyst. Thus, although with $AlCl_3$ 0.1 M, the xylose conversion was higher, 97 % *vs* 78.3 %, this did not lead to a significant improvement in the furfural production, 4 g/L *vs* 3.5 g/L. In other experiments carried out at 200 °C with a higher catalyst concentration, the xylose was almost completely converted, and conversion rates of approximately 97 % were determined (runs 7, 8), but in these cases, the furfural production did not improve either. This can be attributed to the furfural loss and the formation of by-products like formic acid when the catalyst was used. A previous work reported that chloride salts improved the reaction of xylose to furfural in an acid medium at 170 – 200 °C (De Jong and Marcotullio, 2010). However, in the present work, the use of a higher salt concentration caused a drop in the furfural yield (Table 5). This was attributed to the harsh treatment, which caused furfural degradation and formic acid formation. (Brazdausks et al., 2022a) In this work, the initial formic acid concentration in the liquor, 1.20 ± 0.10 g/L, was increased up to 4.3 g/L when the treatment was performed at 200 °C with $AlCl_3$ concentrations of 0.2 M (run 7) and 0.3 M (run 8). To overcome this inconvenience, biphasic systems may provide an interesting approach to the conversion of xylose to furfural by avoiding degradation of the formed product. For example, an iron-catalyzed furfural production in a biphasic system H_2O/2-MeTHF, resulting in effective dehydration in the aqueous phase, and 98 % of the furfural was in situ extracted into the 2-MeTHF. (Vomstein et al., 2011) The results obtained in this work are comparable to those obtained from corn stover treated at 210 °C for 20 min without a catalyst, which provided a maximum furfural yield of 46.5 % (Li et al., 2020), or from birch wood chips treated at 175 °C for 90 min with H_3PO_4 as the catalyst, with yields of 63 – 72 % (Brazdausks et al., 2022b). Rivas et al., (2021) reported a maximum furfural yield of 64 %

Table 5
Chemical conversions of xylose to furfural under different experimental conditions.

Run	$AlCl_3$ concentration (M)	Residual xylose concentration (g/L)	Formic acid concentration (g/L)	Furfural concentration (g/L)	Furfural yield (%)	Conversion (%)
			170 °C			
1	0.0	29.30 ± 0.13	2.74 ± 0.02	4.14 ± 0.11	20.76	6.08
2	0.1	20.24 ± 0.09	2.77 ± 0.10	4.91 ± 0.16	24.58	35.10
3	0.2	14.62 ± 0.24	2.93 ± 0.04	7.86 ± 0.03	39.36	53.14
4	0.3	9.99 ± 0·23	3.16 ± 0.07	9.88 ± 0.27	49.50	67.99
			200 °C			
5	0.0	6.78 ± 0.23	3.48 ± 0.09	12.49 ± 0.02	62.55	78.26
6	0.1	0.97 ± 0.15	4.03 ± 0.07	12.80 ± 0.03	64.10	96.88
7	0.2	0.79 ± 0.08	4.32 ± 0.05	11.13 ± 0.16	55.75	97.48
8	0.3	0.86 ± 0.10	4.25 ± 0.03	9.44 ± 0.18	47.31	97.26

Median concentrations from three replicates and the standard deviations.

from vine shoots pretreated in water/1-butanol/ H_2SO_4 media at 170 ºC using a microwave reactor (Rivas et al., 2021). Previous works have optimized the furfural production in a microwave reactor at 200 ºC using $FeCl_3$ as a catalyst from vine shoots with a furfural yield of 73 % (Castro et al., 2023) and from olive stones with a yield of 63.3 %. (Padilla-Rascón et al., 2021)

4. Conclusions

We have shown that the OrganoCat treatment can be employed in an integrated approach for the holistic use of relatively unexplored vine shoot lignocellulosic biomass for the production of several targeted bio-based products. The treatment effectively separated the components, yielding a cellulose-rich pulp, hydrolyzed hemicelluloses, and high-purity lignin. The cellulose-rich solid is efficiently used in the ethanol production via the PSSF process, achieving approximately 70 % of the theoretical yield and final ethanol concentrations close to 33 g/L The aqueous phase with sugars in dissolution was viable sources for ethanol and furfural, with conversion values reaching up to 98 % and 64 %, respectively. The process also recovered 41.3 % of the initial lignin with high purity. Overall, the OrganoCat method is a promising pretreatment for the fractionation and harnessing of all of the fractions for vine shoots' lignocellulosic biomass.

CRediT authorship contribution statement

Silvia Morales-delaRosa: Writing – review & editing, Supervision, Project administration, Methodology, Investigation, Formal analysis, Conceptualization. **Marta Lara-Serrano:** Writing – original draft, Methodology, Investigation. **Paloma Manzanares:** Writing – review & editing, Writing – original draft, Supervision, Project administration, Funding acquisition, Conceptualization. **Jose M Oliva:** Writing – review & editing, Writing – original draft, Resources, Methodology, Investigation. **Eulogio Castro:** Writing – review & editing, Resources, Project administration, Formal analysis, Conceptualization. **Inmaculada Romero:** Writing – review & editing, Writing – original draft, Resources, Project administration, Methodology, Investigation, Conceptualization. **Campos-Martin Jose M:** Writing – review & editing, Validation, Supervision, Resources, Funding acquisition, Conceptualization.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.indcrop.2025.121240.

Data availability

Data will be made available on request.

References

Basak, B., Kumar, R., Sai Bharadwa, A.V.S.L., Kim, T.H., Kim, J.R., Jang, M., Oh, S.E., Roh, H.S., Jeon, B.H., 2023. Advances in physicochemical pretreatment strategies for lignocellulose biomass and their effectiveness in bioconversion for biofuel production. Bioresour. Technol. 369, 128413. https://doi.org/10.1016/j.biortech.2022.128413.

Bozell, J.J., Petersen, G.R., 2010. Technology development for the production of biobased products from biorefinery carbohydrates—the US Department of Energy's "top 10" revisited. Green. Chem. 12, 539–555. https://doi.org/10.1039/b922014c.

Brazdausks, P., Godina, D., Puke, M., 2022a. Direct furfural production from deciduous wood pentosans using different phosphorus-containing catalysts in the context of biorefining. Molecules 27, 7353. https://doi.org/10.3390/molecules27217353.

Brazdausks, P., Godina, D., Puke, M., 2022b. Direct furfural production from deciduous wood pentosans using different phosphorus-containing catalysts in the context of biorefining. Molecules 27, 7353. https://doi.org/10.3390/molecules27217353.

Cai, T., Liu, C., Jiang, J., Meng, X., Ragauskas, A.J., Wang, K., 2024. Innovative biphasic solvent systems for lignocellulosic biorefinery. Trends Chem. 6, 219–233. https://doi.org/10.1016/j.trechm.2024.03.003.

Cardoza, D., Contreras, M. del M., Lara-Serrano, M., Morales-delaRosa, S., Campos-Martín, J.M., Romero, I., Castro, E., 2024. Sustainable vine shoots-to-ethanol valorisation by a sequential acid/organosolv pretreatment. Process Saf. Environ. Prot. 183, 1059–1070. https://doi.org/10.1016/j.psep.2024.01.063.

Castro, E., Rabelo, C.A.B.S., Padilla-Rascón, C., Vidal, A.M., López-Linares, J.C., Varesche, M.B.A., Romero, I., 2023. Biological hydrogen and furfural production from steam-exploded vine shoots. Renew. Energy 219, 119393. https://doi.org/10.1016/j.renene.2023.119393.

Cebrián-Tarancón, C., Fernández-Roldán, F., Sánchez-Gómez, R., Salinas, R., Llorens, S., 2021. Vine-shoots as enological additives. A study of acute toxicity and cytotoxicity. Foods 10, 1267. https://doi.org/10.3390/foods10061267.

Contreras, M. del M., Romero-García, J.M., López-Linares, J.C., Romero, I., Castro, E., 2022. Residues from grapevine and wine production as feedstock for a biorefinery. Food Bioprod. Process 134, 56–79. https://doi.org/10.1016/j.fbp.2022.05.005.

Dávila, I., Gullón, P., Andrés, M.A., Labidi, J., 2017. Coproduction of lignin and glucose from vine shoots by eco-friendly strategies: Toward the development of an integrated biorefinery. Bioresour. Technol. 244, 328–337. https://doi.org/10.1016/j.biortech.2017.07.104.

De Jong, W., Marcotullio, G., 2010. Overview of biorefineries based on co-production of furfural, existing concepts and novel developments. Int. J. Chem. React. Eng. 8. https://doi.org/10.2202/1542-6580.2174.

Djellabi, R., Aboagye, D., Galloni, M.G., Vilas Andhalkar, V., Nouacer, S., Nabgan, W., Rtimi, S., Constantí, M., Medina Cabello, F., Contreras, S., 2023. Combined conversion of lignocellulosic biomass into high-value products with ultrasonic cavitation and photocatalytic produced reactive oxygen species – a review. Bioresour. Technol. 368, 128333. https://doi.org/10.1016/j.biortech.2022.128333.

Ershova, O., Nieminen, K., Sixta, H., 2017. The role of various chlorides on xylose conversion to furfural: experiments and kinetic modeling. ChemCatChem 9, 3031–3040. https://doi.org/10.1002/cctc.201700269.

Fernández-Rodríguez, J., Erdocia, X., Sánchez, C., González Alriols, M., Labidi, J., 2017. Lignin depolymerization for phenolic monomers production by sustainable processes. J. Energy Chem. 26, 622–631. https://doi.org/10.1016/j.jechem.2017.02.007.

French, A.D., 2014. Idealized powder diffraction patterns for cellulose polymorphs. Cellulose 21, 885–896. https://doi.org/10.1007/s10570-013-0030-4.

French, A.D., Santiago Cintrón, M., 2013. Cellulose polymorphy, crystallite size, and the Segal Crystallinity Index. Cellulose 20, 583–588. https://doi.org/10.1007/s10570-012-9833-y.

Gao, B., Sun, C., Yang, T., Wen, Q., You, S., Yang, Q., Yang, Z., Cheng, H., Wang, Y., Zhou, H., Chen, Z., 2023. Biphasic solvent systems enabled lignocellulosic biomass fractionation: A pathway towards comprehensive biomass utilization. Ind. Crops Prod. 202, 117036. https://doi.org/10.1016/j.indcrop.2023.117036.

Grande, P.M., Schoofs, L., Weidener, D., Leitner, W., Klose, H., 2024. Lignocellulose treatment using a flow-through variant of organocat process. ChemSusChem 18, e202401063. https://doi.org/10.1002/cssc.202401063.

Grande, P.M., Viell, J., Theyssen, N., Marquardt, W., Domínguez De María, P., Leitner, W., 2015. Fractionation of lignocellulosic biomass using the OrganoCat process. Green. Chem. 17, 3533–3539. https://doi.org/10.1039/c4gc02534b.

Hergert, H.L., 1960. Infrared spectra of lignin and related compounds. II. Conifer lignin and model compounds1,2. J. Org. Chem. 25, 405–413. https://doi.org/10.1021/jo01073a026.

Holechek, J.L., Geli, H.M.E., Sawalhah, M.N., Valdez, R., 2022. A global assessment: can renewable energy replace fossil fuels by 2050? Sustainability 14. https://doi.org/10.3390/su14084792.

Hu, M., Yu, Y., Liu, Y., 2023. Integrated strategies for furfural production and lignocellulose fractionation in aqueous deep eutectic solvents. Ind. Crops Prod. 204, 117334. https://doi.org/10.1016/J.INDCROP.2023.117334.

IEA, 2023. Fossil Fuels Consumption Subsidies 2022, ⟨https://www.iea.org/reports/fossil-fuels-consumption-subsidies-2022⟩, Elgar Encyclopedia of Climate Policy.

Kapu, N.S., Trajano, H.L., 2014. Review of hemicellulose hydrolysis in softwoods and bamboo. Biofuels, Bioprod. Bioref. 8, 857–870. https://doi.org/10.1002/bbb.1517.

Kassim, M.A., Meng, T.K., Serri, N.A., Yusoff, S.B., Shahrin, N.A.M., Seng, K.Y., Bakar, M. H.A., Keong, L.C., 2020. Sustainable biorefinery concept for industrial bioprocessing. : Biorefinery Prod. Technol. Chem. Energy 15–53. https://doi.org/10.1002/9781119593065.ch2.

Koppram, R., Tomás-Pejó, E., Xiros, C., Olsson, L., 2014. Lignocellulosic ethanol production at high-gravity: challenges and perspectives. Trends Biotechnol. 32, 46–53. https://doi.org/10.1016/j.tibtech.2013.10.003.

Langletz, T., Grande, P.M., Viell, J., Wolters, D., Tonn, J., Klose, H., Schriever, S.G., Hoffmann, A., Jupke, A., Gries, T., Herres-Pawlis, S., 2024. Toward a greener bioeconomy: synthesis and characterization of lignin–polylactide copolymers. Adv. Energy Sustain. Res. 5. https://doi.org/10.1002/aesr.202300187.

Lara-Serrano, M., Fierro, J.L.G., Morales-Delarosa, S., Campos-Martín, J.M., 2020. High enhancement of the hydrolysis rate of cellulose after pretreatment with inorganic salt hydrates. Green Chem. 22, 3860–3866. https://doi.org/10.1039/d0gc01066a.

Lara-Serrano, M., Morales-delaRosa, S., Campos-Martín, J.M., Fierro, J.L.G., 2019. Fractionation of lignocellulosic biomass by selective precipitation from ionic liquid dissolution. Appl. Sci. 9, 1862. https://doi.org/10.3390/app9091862.

Lara-Serrano, M., Sboiu, D.M., Morales-delaRosa, S., Campos-Martin, J.M., 2023. Selective fragmentation of lignocellulosic biomass with ZnCl2·4H2O using a dissolution/precipitation method. Appl. Sci. 13, 2953. https://doi.org/10.3390/app13052953.

Li, W.C., Zhang, S.J., Xu, T., Sun, M.Q., Zhu, J.Q., Zhong, C., Li, B.Z., Yuan, Y.J., 2020. Fractionation of corn stover by two-step pretreatment for production of ethanol, furfural, and lignin. Energy 195, 117076. https://doi.org/10.1016/J.ENERGY.2020.117076.

López-Linares, J.C., Romero, I., Cara, C., Ruiz, E., Moya, M., Castro, E., 2014. Bioethanol production from rapeseed straw at high solids loading with different process configurations. Fuel 122, 112–118. https://doi.org/10.1016/j.fuel.2014.01.024.

Mankar, A.R., Pandey, A., Modak, A., Pant, K.K., 2021. Pretreatment of lignocellulosic biomass: a review on recent advances. Bioresour. Technol. 334, 125235. https://doi.org/10.1016/j.biortech.2021.125235.

Mariscal, R., Maireles-Torres, P., Ojeda, M., Sádaba, I., López Granados, M., 2016. Furfural: a renewable and versatile platform molecule for the synthesis of chemicals and fuels. Energy Environ. Sci. 9, 1144–1189. https://doi.org/10.1039/c5ee02666k.

Martinez Diaz, J., Grande, P.M., Klose, H., 2024. Examining poplar biomass recalcitrance in OrganoCat processing: the impact of genotype, nitrogen fertilization and process severity. Ind. Crop. Prod. 222, 119820. https://doi.org/10.1016/j.indcrop.2024.119820.

Morales-delaRosa, S., Campos-Martin, J.M., Fierro, J.L.G., 2014. Complete chemical hydrolysis of cellulose into fermentable sugars through ionic liquids and antisolvent pretreatments. ChemSusChem 7, 3467–3475. https://doi.org/10.1002/cssc.201402466.

Moreno, A.D., Duque, A., González, A., Ballesteros, I., Negro, M.J., 2021. Valorization of greenhouse horticulture waste from a biorefinery perspective. Foods 10, 814. https://doi.org/10.3390/foods10040814.

OIV, 2023. State of the world vine and wine sector in 2022, International Organization of vine and wine.

Padella, M., O'Connell, A., Prussi, M., 2019. What is still limiting the deployment of cellulosic ethanol? Analysis of the current status of the sector. Appl. Sci. 9, 4523. https://doi.org/10.3390/app9214523.

Padilla-Rascón, C., Romero-García, J.M., Ruiz, E., Castro, E., 2020. Optimization with response surface methodology of microwave-assisted conversion of xylose to furfural. Molecules 25, 3574. https://doi.org/10.3390/molecules25163574.

Padilla-Rascón, C., Romero-García, J.M., Ruiz, E., Romero, I., Castro, E., 2021. Microwave-assisted production of furfural from the hemicellulosic fraction of olive stones. Process Saf. Environ. Prot. 152, 630–640. https://doi.org/10.1016/j.psep.2021.06.035.

Palacios, A.S., Ilyina, A., Ramos-González, R., Aguilar, C.N., Martínez-Hernández, J.L., Segura-Ceniceros, E.P., González, M.L.C., Aguilar, M., Ballesteros, M., Oliva, J.M., Ruiz, H.A., 2021. Ethanol production from banana peels at high pretreated substrate loading: comparison of two operational strategies. Biomass-.-. Convers. Biorefinery 11, 1587–1596. https://doi.org/10.1007/s13399-019-00562-7.

Pandey, K.K., Pitman, A.J., 2003. FTIR studies of the changes in wood chemistry following decay by brown-rot and white-rot fungi. Int. Biodeterior. Biodegrad 52, 151–160. https://doi.org/10.1016/S0964-8305(03)00052-0.

Park, J.H., Kim, S.H., Park, H.D., Kim, J.S., Yoon, J.J., 2014. Simultaneous utilization of galactose and glucose by Saccharomyces cerevisiae mutant strain for ethanol production. Renew. Energy 65, 213–218. https://doi.org/10.1016/j.renene.2013.09.010.

Patel, A., Shah, A.R., 2021. Integrated lignocellulosic biorefinery: Gateway for production of second generation ethanol and value added products. J. Bioresour. Bioprod. 6, 108–128. https://doi.org/10.1016/j.jobab.2021.02.001.

Prasad, B.R., Padhi, R.K., Ghosh, G., 2023. A review on key pretreatment approaches for lignocellulosic biomass to produce biofuel and value-added products. Int. J. Environ. Sci. Technol. 20, 6929–6944. https://doi.org/10.1007/s13762-022-04252-2.

Raj, T., Chandrasekhar, K., Naresh Kumar, A., Rajesh Banu, J., Yoon, J.J., Kant Bhatia, S., Yang, Y.H., Varjani, S., Kim, S.H., 2022. Recent advances in commercial biorefineries for lignocellulosic ethanol production: Current status, challenges and future perspectives. Bioresour. Technol. 344, 126292. https://doi.org/10.1016/j.biortech.2021.126292.

Rivas, S., López, L., Vila, C., Parajó, J.C., 2021. Organosolv processing of vine shoots: Fractionation and conversion of hemicellulosic sugars into platform chemicals by microwave irradiation. Bioresour. Technol. 342. https://doi.org/10.1016/j.biortech.2021.125967.

Romero-García, J.M., Susmozas, A., Padilla-Rascón, C., Manzanares, P., Castro, E., Oliva, J.M., Romero, I., 2022. Ethanol production from olive stones using different process strategies. Renew. Energy 194, 1174–1183. https://doi.org/10.1016/j.renene.2022.06.022.

Roy, S., Chundawat, S.P.S., 2023. Ionic liquid–based pretreatment of lignocellulosic biomass for bioconversion: a critical review. Bioenergy Res. 16, 263–278. https://doi.org/10.1007/s12155-022-10425-1.

Sai Bharadwaj, A.V.S.L., Dev, S., Zhuang, J., Wang, Y., Yoo, C.G., Jeon, B.H., Aggarwal, S., Park, S.H., Kim, T.H., 2023. Review of chemical pretreatment of lignocellulosic biomass using low-liquid and low-chemical catalysts for effective bioconversion. Bioresour. Technol. 368, 128339. https://doi.org/10.1016/j.biortech.2022.128339.

Schoofs, L., Thiele, B., Tonn, J., Langletz, T., Herres-Pawlis, S., Jupke, A., Grande, P.M., Klose, H., 2024. Structural changes of lignin during organocat processing. Energy Fuels 38, 4192–4202. https://doi.org/10.1021/acs.energyfuels.3c03673.

Schoofs, L., Weidener, D., Schurr, U., Klose, H., Grande, P.M., 2021. Fractionation of lignocellulosic biomass using the organocat process. J. Vis. Exp. 2021. https://doi.org/10.3791/61933.

Schwiderski, M., Kruse, A., Grandl, R., Dockendorf, D., 2014. Comparison of the influence of a Lewis acid AlCl3 and a Brønsted acid HCl on the organosolv pulping of beech wood. Green Chem. 16, 1569–1578. https://doi.org/10.1039/c3gc42050g.

Singh, P., Kiran, U., Ghosh, S., 2025. Repeated batch fermentation of xylose-rich hydrolysate obtained from wheat straw in a bioreactor to produce second-generation bioethanol. Fuel 387, 134383. https://doi.org/10.1016/j.fuel.2025.134383.

Sluiter, A., Hames, B., Ruiz, R.O., Scarlata, C., Sluiter, J., Templeton, D., Energy, D. of, 2004. Determination of structural carbohydrates and lignin in biomass. Biomass Anal. Technol. Team Lab. Anal. Proced. 1–14.

Sluiter, J.B., Ruiz, R.O., Scarlata, C.J., Sluiter, A.D., Templeton, D.W., 2010. Compositional analysis of lignocellulosic feedstocks. 1. Review and description of methods. J. Agric. Food Chem. 58, 9043–9053. https://doi.org/10.1021/jf1008023.

Sun, R.C., Fang, J.M., Tomkinson, J., Geng, Z.C., Liu, J.C., 2001. Fractional isolation, physico-chemical characterization and homogeneous esterification of hemicelluloses from fast-growing poplar wood. Carbohydr. Polym. 44, 29–39. https://doi.org/10.1016/S0144-8617(00)00196-X.

Sun, X.F., Xu, F., Sun, R.C., Geng, Z.C., Fowler, P., Baird, M.S., 2005. Characteristics of degraded hemicellulosic polymers obtained from steam exploded wheat straw. Carbohydr. Polym. 60, 15–26. https://doi.org/10.1016/j.carbpol.2004.11.012.

Vomstein, T., Grande, P.M., Leitner, W., Domínguezdemaría, P., 2011. Iron-catalyzed furfural production in biobased biphasic systems: From pure sugars to direct use of crude xylose effluents as feedstock. ChemSusChem 4, 1592–1594. https://doi.org/10.1002/cssc.201100259.

Wang, J., Azam, W., 2024. Natural resource scarcity, fossil fuel energy consumption, and total greenhouse gas emissions in top emitting countries. Geosci. Front 15, 101757. https://doi.org/10.1016/j.gsf.2023.101757.

Wang, Fang, Harindintwali, J.D., Yuan, Zhizhang, Wang, M., Wang, Faming, Li, S., Yin, Z., Huang, L., Fu, Y., Li, L., Chang, S.X., Zhang, L., Rinklebe, J., Yuan, Zuoqiang, Zhu, Q., Xiang, L., Tsang, D.C.W., Xu, L., Jiang, X., Liu, J., Wei, N., Kästner, M., Zou, Y., Ok, Y.S., Shen, J., Peng, D., Zhang, W., Barceló, D., Zhou, Y., Bai, Z., Li, B., Zhang, B., Wei, K., Cao, H., Tan, Z., Zhao, L. bin, He, X., Zheng, J., Bolan, N., Liu, X., Huang, C., Dietmann, S., Luo, M., Sun, N., Gong, J., Gong, Y., Brahushi, F., Zhang, T., Xiao, C., Li, X., Chen, W., Jiao, N., Lehmann, J., Zhu, Y.G., Jin, H., Schäffer, A., Tiedje, J.M., Chen, J.M., 2021. Technologies and perspectives for achieving carbon neutrality. Innovation 2, 100180. https://doi.org/10.1016/j.xinn.2021.100180.

Wang, X., Hu, M., Liu, Y., 2025. Efficient separation of lignocellulose component and furfural production from hemicellulose using a γ-valerolactone/H2O system. Int. J. Biol. Macromol. 311, 143749. https://doi.org/10.1016/J.IJBIOMAC.2025.143749.

Xie, X., Li, L., Wang, X., Liu, Q., Ouyang, X., Qiu, X., 2025. Fractionation of high-yield noncondensed lignin and glucan oligomers from lignocellulose in a novel biphasic system. J. Agric. Food Chem. 73, 2880–2889. https://doi.org/10.1021/acs.jafc.4c12533.

Yan, Q., Miazek, K., Grande, P.M., Domínguez De María, P., Leitner, W., Modigell, M., 2014. Mechanical pretreatment in a screw press affecting chemical pulping of lignocellulosic biomass. Energy Fuels 28, 6981–6987. https://doi.org/10.1021/ef501706w.

Yogalakshmi, K.N., Poornima Devi, T., Sivashanmugam, P., Kavitha, S., Yukesh Kannah, R., Sunita, V., AdishKumar, S., Gopalakrishnan, K., Rajesh Banu, J., 2022. Lignocellulosic biomass-based pyrolysis: a comprehensive review. Chemosphere 286, 131824. https://doi.org/10.1016/j.chemosphere.2021.131824.

Zhang, M., Wang, F., Su, R., Qi, W., He, Z., 2010. Ethanol production from high dry matter corncob using fed-batch simultaneous saccharification and fermentation after combined pretreatment. Bioresour. Technol. 101, 4959–4964. https://doi.org/10.1016/j.biortech.2009.11.010.

Zhao, X., Dong, L., Chen, L., Liu, D., 2013. Batch and multi-step fed-batch enzymatic saccharification of Formiline-pretreated sugarcane bagasse at high solid loadings for high sugar and ethanol titers. Bioresour. Technol. 135, 350–356. https://doi.org/10.1016/j.biortech.2012.09.074.

Zhou, Z., Ouyang, D., Liu, D., Zhao, X., 2023. Oxidative pretreatment of lignocellulosic biomass for enzymatic hydrolysis: progress and challenges. Bioresour. Technol. 367, 128208. https://doi.org/10.1016/j.biortech.2022.128208.

Zhou, M., Tian, X., 2022. Development of different pretreatments and related technologies for efficient biomass conversion of lignocellulose. Int. J. Biol. Macromol. 202, 256–268. https://doi.org/10.1016/j.ijbiomac.2022.01.036.