

# Biovalorisation and Sustainability of Agrofood Products

## Lignocellulosic Biorefinery

### Lignin Oxidation

**KEYWORDS:** Biorefinery / Lignin / Depolymerization / Oxidation / Phenolic monomers / Dicarboxylic acids

This research topic includes an overview of the main contributions of lignin valorization through oxidative depolymerization to produce added-value compounds developed over the recent years in LSRE-LCM. An evaluation of the valuable products, such as aldehydes (vanillin and syringaldehyde) and acids (vanillin acid and syringic acid), obtained from oxidation in an alkaline medium with the oxygen of lignins and liquors from different sources, delignification processes, and oxidation conditions is provided. Lignin oxidative depolymerization towards C<sub>4</sub>-dicarboxylic acids, like succinic and oxalic acids, was also studied, emphasizing catalytic conversion by H<sub>2</sub>O<sub>2</sub> oxidation.

#### Introduction

Lignin is the second most abundant component, next to cellulose, in lignocellulosic biomass. Large amounts of this polymer are produced annually in the pulp and paper industries as a coproduct from the cooking process - most of it burned as fuel for energy. Strategies regarding lignin valorization have attracted significant attention due to lignin's aromatic structure. Oxidative depolymerization allows the conversion of lignin into added-value compounds, such as phenolic monomers and/or dicarboxylic acids, which could be an excellent alternative to aromatic petrochemicals. Despite all the depolymerization methods, oxidation in an alkaline medium using oxygen aroused great interest in lignin valorization since it represents an effective method for added-value compound production from different sources of lignin. Oxidative conversion gives a complex mixture of products highly dependent on the nature of the raw material and the selected reaction conditions. Among them, it is possible to find oligomeric products, non-phenolic, and phenolic compounds, as vanillin and syringaldehyde.

Besides the wide application of oxidation in an alkaline medium with oxygen for lignin depolymerization to phenolic compounds, this method is less effective towards C<sub>4</sub>-dicarboxylic acids. Oxidation products, like adipic, muconic, succinic, maleic, and oxalic acids, could be obtained from lignin catalytic wet peroxide oxidation since this method allows the cleavage of the aromatic ring in the lignin structure. Although, the product selectivity depends on the oxidizing agent, catalyst type, and process conditions.

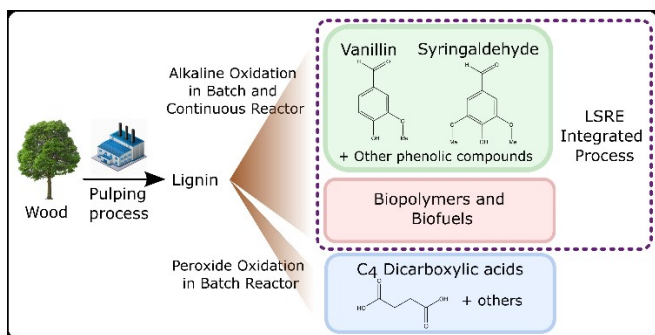


Fig 1. Processes of lignin oxidative depolymerization studied in LSRE-LCM.

#### Current Development

Alkaline oxidation converts lignin into a complex mixture of products that could be phenolic monomers, dimers, and oligomers. Since the selectivity and efficiency of oxidative depolymerization depend strongly on processing conditions and lignin origin, the effect of one or several process parameters on lignin oxidative depolymerization performed through batch experiments has been investigated. The primary objective was to achieve the ideal conditions for obtaining the maximum conversion of lignin while avoiding the oxidation of the phenolic monomers produced. The origin, composition, and processing of lignin (pulping process, isolation method, pretreatment, etc.), the oxygen partial pressure, the initial temperature, and the lignin and sodium hydroxide concentration in the reaction solution were studied, and its effect on the selectivity and efficiency of alkaline oxidative depolymerization process was evaluated.

It was found that high partial pressure of oxygen reduces reaction time but leads to an increased rate of vanillin oxidation. Increasing the oxygen pressure accelerated both product formation and degradation, and therefore, shortened the time needed to reach the maximum product yields. What concerns the sodium hydroxide concentration, the effect of the pH of the mixture, it was concluded that during the lignin oxidation process, the yield of vanillin decreased when the pH value began to decrease. Moreover, there was a smaller vanillin degradation for strong alkaline conditions that increased significantly when pH was smaller than 11.5. Consequently, high alkali concentrations (pH>12) are needed to reduce vanillin degradation. For temperature, it was found that an increase in this reaction condition can shorten the reaction time but, on the other hand, result in faster degradation of aldehydes produced. Finally, the lignin itself is a variable with a huge influence on the final yields of oxidation products. It was found that vanillin yield only increased for lignin concentrations up to 60 g/L, decreasing for higher values.

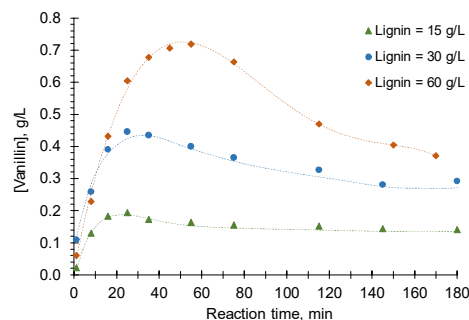


Fig 2. Effect of initial lignin concentration (15, 30, 60 g/L) on vanillin concentration during oxidation. General conditions: T<sub>initial</sub> = 393 K, pH<sub>initial</sub> ≥ 13.8, P<sub>total</sub> = 9.8 bar, pO<sub>2</sub> = 3 bar.

As already stated, during oxidation the formation of the main phenolic products can be simultaneously accompanied by their degradation process, which is dependent on the applied oxidation conditions. For this reason, the individual degradation under oxidation conditions mimicking lignin oxidation is an important tool to better understand this reaction and maximize the yield of target value-added products. The effect of temperature, phenolic monomers initial

concentration, and oxygen partial pressure was evaluated, and a simple mathematical model was developed to describe the data from the degradation of vanillin, vanillic acid, acetovanillone, syringaldehyde, syringic acid, and acetosyringone during oxidation reactions.

The obtained data showed that the initial concentration of phenolic compounds as well as the oxygen concentration did not affect the degradation reaction rate, since a first-order dependence for both oxidation parameters was obtained for all the studied compounds.

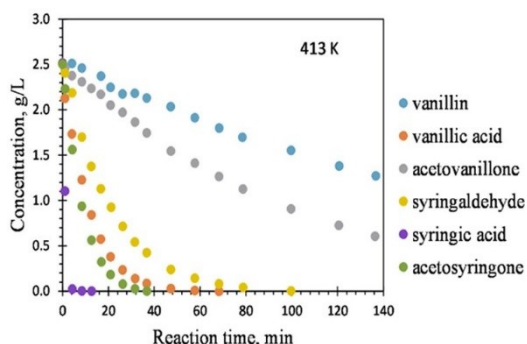


Fig 3. Phenolic monomers degradation as a function of oxidation reaction time. Initial reaction conditions: [NaOH] = 80 g/L;  $T_{\text{initial}} = 413 \text{ K}$ ,  $\text{pH}_{\text{initial}} \approx 14$ ; phenolics initial concentration = 2.5 g/L;  $\text{pO}_2 = 3.0 \text{ bar}$ ;  $\text{P}_{\text{total}} = 9.8 \text{ bar}$ .

Comparing all the results of the studied phenolic, it was possible to conclude that syringaldehyde, syringic acid, and acetosyringone have higher degradation rates in comparison with vanillin, vanillic acid, and acetovanillone. This is an expected behavior since derivatives from S units are more reactive than the ones from G due to the presence of two methoxyl groups in the aromatic ring in the later compounds.

The study of peroxide oxidation aims to obtain dicarboxylic acids (DCA), focused on  $\text{C}_4$ -DCA (mainly succinic, malic, fumaric, maleic, and tartaric acids), and specifically succinic acid. Several of these dicarboxylic acids have shown relevance as feedstocks for polymers, cosmetics, and the food industry. Considering peroxide oxidation, it has been stated that this oxidant is considered more efficient for aromatic ring cleavage than oxygen used in oxidation in alkaline medium. Moreover, the peroxide releases free radicals, which enhance lignin degradation in a shorter time under less severe conditions.

The oxidation of vanillic acid, a lignin model compound, was studied under catalytic wet peroxide oxidation conditions, using titanium silicalite-1 (TS-1) as the catalyst. The effects of temperature, pH, and reaction time were studied. In the second phase, catalyst modification with transition metal oxides (Fe, Co, Cu) was also tested.

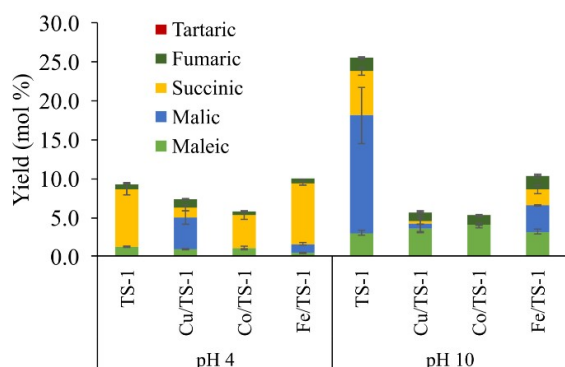


Fig 4. Effect of titanium silicalite-1 (TS-1) and modified TS-1 catalysts in the  $\text{C}_4$ -DCA yields for the wet peroxide oxidation of vanillic acid.

The use of a TS-1 catalyst in the catalytic wet peroxide oxidation enhanced the production of  $\text{C}_4$ -DCA, comparatively with the non-catalyzed reaction. The type of produced acids depends mainly on the used temperature, pH, and reaction time. An alkaline medium caused an over-oxidation of vanillic acid, giving rise to hydroxylated acids, like malic and tartaric, together with high degradation levels. In acidic pH, the main produced acid was succinic, and a lower amount of degradation products was observed. Therefore, vanillic acid oxidation acted similarly to other lignin model compounds, where the oxidation of the aromatic ring is the first step, getting open to produce  $\text{C}_4$ -DCA. However, these  $\text{C}_4$ -DCA can quickly degrade to low-molecular-weight compounds, especially under alkaline conditions and long reaction times. If succinic acid is the target compound, acidic oxidation should be selected.

International collaborations:

- The supply of services with Sappi comprised the structural characterization and oxidation of four lignosulfonates. The lignin samples were subjected to nitrobenzene oxidation and oxidation with oxygen in an alkaline medium and also analyzed by  $^{13}\text{C}$  NMR.
- The work developed in the supply of services with Bloom Biorenewables included oxidative depolymerization of a provided uncondensed lignin through nitrobenzene oxidation and oxidation with oxygen in an alkaline medium with different initial reaction conditions.

#### Future Perspectives

Progress/future work within this research topic includes:

- performance evaluation of lignin oxidation experiments to find the best reaction conditions that enable the improvement of phenolic products yields depending on the lignin sample;
- identification of new added-value compounds from lignin;
- kinetic study of all the phenolic products formed from lignin oxidation with and without catalysts;
- development of a staged membrane process to fractionate the oxidation mixture and valorize the fractions produced;
- study of the obtained oxidized lignin fraction(s) with higher molecular weight to produce new products.

#### Related Sustainable Development Goals



#### Outputs

##### PhD Theses

- [1] Elson Gomes, Development of a continuous process for the production of vanillin and syringaldehyde from kraft black liquor, PDEQB, FEUP, 2019
- [2] Carlos Vega Aguilar, Production of  $\text{C}_4$  dicarboxylic acids from lignin oxidation, PDEQB, FEUP, 2021
- [3] Filipa Casimiro, Lignin from pulp mill sulfite liquors: Production of vanillin and syringaldehyde by an integrated process within the biorefinery concept, PDEQB, FEUP, 2023

##### Selected Publications

- [1] F.M. Casimiro et al., *Ind. Eng. Chem. Res.* 58, 16442 (2019)
- [2] C. Vega Aguilar et al., *Ind. Eng. Chem. Res.* 60, 3543 (2021)
- [3] C.A.E. Costa et al., *Molecules* 26, 4602 (2021)
- [4] C. Vega Aguilar et al., *Ind. Crops and Products* 173, 114155 (2021)
- [5] C.A.E. Costa et al., *ChemEngineering*, 7, 42, (2023)

##### Team

**Alírio Rodrigues**, Professor; **Carina Costa**, Researcher; **Elson Gomes**, PhD Student; **Filipa Casimiro**, PhD Student; **Carlos Vega**, PhD Student

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LSRE-LCM Base Funding, UIDB/50020/2020, 2020-2023  
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# Biovalorisation and Sustainability of Agrofood Products

## Lignocellulosic Biorefinery

### Fractionation and Purification of Syringaldehyde and Vanillin

**KEYWORDS:** Biorefinery / Lignin / Phenolic compounds / Membrane filtration / Adsorption / Crystallization

In this research topic, the fractionation and purification of added-value phenolic compounds, produced by the alkaline oxidation of lignin, is discussed. A sequence of physical separation processes was assessed for the recovery of vanillin and syringaldehyde from an oxidatively depolymerized lignin solution. Ultra- and nanofiltration membranes were used to reduce the complexity of the oxidized lignin mixture by separating the high molecular-weight fractions of degraded lignin from the lower molecular-weight species and then, the alkaline permeate from nanofiltration was submitted to adsorption using a polymeric resin. Two desorption eluents were applied during the adsorption cycles, deionized water and ethanol. The filtration and adsorption/desorption sequence allows the separation and purification of selected compounds in dilute solutions that proceed to the crystallization process.

#### Introduction

In the concept of biorefineries, the production of added-value compounds from lignin depolymerization is still hindered by the complexity of separation and purification processes needed to obtain the purified phenolic compounds. The reaction solution resulting from oxidative lignin depolymerization generally consists of a complex mixture containing lignin oligomers, low-molecular-weight aromatics, and minor amounts of other secondary non-phenolic compounds. The low molecular-weight aromatics produced are usually difunctional and include aromatic aldehydes (vanillin and syringaldehyde), their respective aromatic acids (vanillic and syringic acids), and ketones (acetovanillone and acetosyringone). The chemical and physical similarities between these phenolic aromatic monomers (their molecular weight, acid dissociation constant, density, and melting point) affect their separation from the oxidized lignin mixture and their consequent purification process. The main sequences of downstream processes used for oxidized lignin mixtures include liquid-liquid extraction, membrane separation, adsorption, ion exchange, distillation, acidification/precipitation, bisulfitation, supercritical fluid extraction, and crystallization. This research topic intended to improve the developed Integrated Process, which includes steps of fractionation and purification of added-value aromatic monomers from a lignin oxidation mixture, such as vanillin and syringaldehyde, prioritizing the low labor- and energy-demanding, and avoiding the use of environmentally harmful solvents, and high material losses.

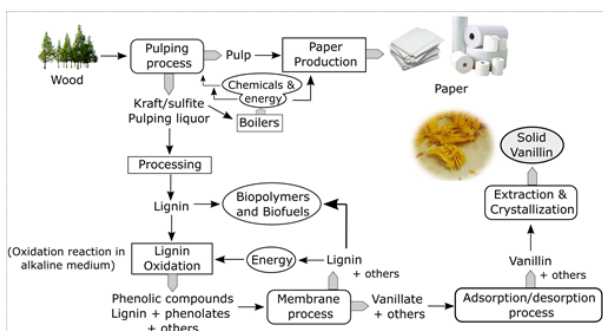


Fig 1. Integrated Process to produce value-added phenolics and polymers from lignin in a biorefinery concept.

#### Current Development

In this topic, valuable information was achieved about the feasibility of the integration of fractionation and purification methods in a sequential process to produce value-added products, such as vanillin and syringaldehyde, from lignin in a biorefinery concept.

A membrane sequence process was applied as a first step of separation to be combined after the production of sodium salts of vanillin and syringaldehyde by alkaline chemical oxidation of lignin. A three-stage membrane of ultra and nanofiltration (molecular weight cut-off of 50, 5, and 1 kDa) was employed and the phenolate content improved during the first membrane fractionation stage. The first membrane stage did not retain the monomers of interest; however, about 9–12 % of the phenolate compounds of interest were retained by the other two membranes and thus, some of the phenolate compounds of interest were lost. One clear advantage of this fractionation sequence employed was the effective decrease in the total non-volatile solids content in the permeate stream, yielding apparent rejection coefficient values between 29 and 15%. Therefore, the choice for pursuing the second and third membrane stages must be carefully evaluated, in the sense that the benefits gained by obtaining a stream containing fewer total solids in the next separation and purification steps must compensate for the loss in the phenolates of interest during membrane processing.

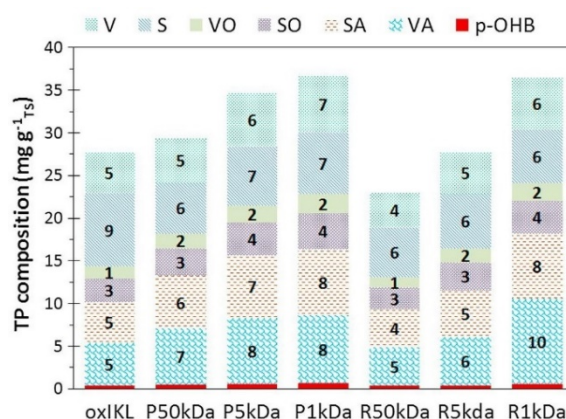


Fig 2. Detailed composition of the phenolate compounds quantified: vanillin (V), syringaldehyde (S), acetovanillone (VO), acetosyringone (SO), vanillic acid (VA), syringic acid (SA) and p-hydroxybenzaldehyde (p-OHB), expressed as mg.g<sup>-1</sup> of total solids (TS) for each processing stream.

The permeate obtained from the nanofractionation process of the oxidized lignin stream was then submitted to adsorption/desorption cycles employing a polymeric resin SP700 without any pH adjustment. The pH of the feed solution influences the separation among aldehydes, acids, and ketones since these phenolic compounds present in oxidized media have different acid dissociation constants (pKa), and therefore can be somewhat tuned by the due to acid dissociation constant differences. The resulting fractions from adsorption/desorption cycles differ considerably from the feed mixture and the monomers are successfully fractionated through their functional groups: phenolic acids, aldehydes, and

ketones. The fraction collected during the feed phase has mostly phenolic acids, such as vanillic acid. Aldehydes were mainly collected in the water desorption phase and the ethanol desorption phase mainly collected the phenolic ketones, such as acetovanillone. Regarding the recovery of each phenolic monomer studied, vanillin showed the highest value, with a recovery higher than 90 %, obtained during the water desorption phase. Vanillic acid, acetovanillone, and syringaldehyde were present in minor quantities, being recovered in the water desorption phase with percentages of about 10, 30, and 60 %, respectively. Moreover, about 40 % and 70 % of the total amount desorbed of syringaldehyde and acetovanillone, respectively, were recovered in the ethanolic desorption phase. Vanillin was also detected in the ethanolic phase but in tracer amounts.

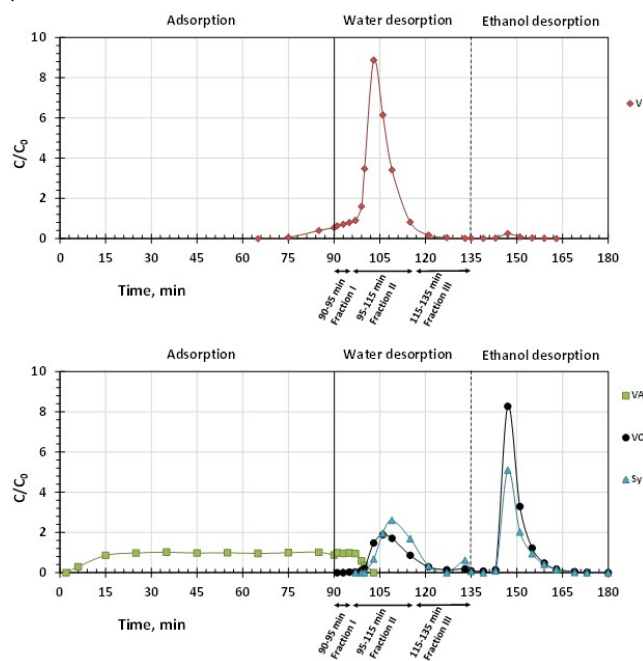


Fig 3. Normalized concentration history of the monomers studied in adsorption/desorption cycles using the permeate from nanofiltration of a softwood lignin oxidized mixture.

The developed work proved that sequential elution with water and ethanol can make possible the separation between families of phenolic compounds present in solution: aldehydes (vanillin and syringaldehyde), acids (vanillic and syringic acids), and ketones (acetovanillone and acetosyringone). This process can simplify the current laborious fractionation and purification of oxidized lignin medium solutions employing several liquid-liquid extractions without harmful solvents and precipitation.

For the first time, the solution obtained from adsorption/desorption experiments applied to an oxidized solution of lignin isolated from softwood sulfite liquor was used for different approaches to cooling and evaporative vanillin crystallization. The influence of the crystallization process on the yield, purity, and polymorphic form of vanillin crystals was evaluated.

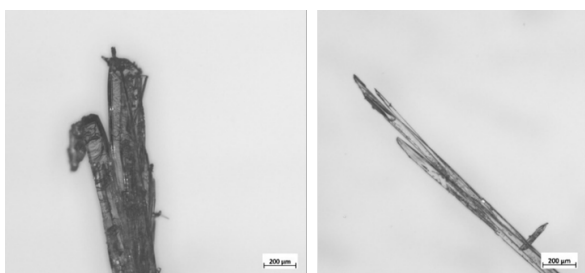


Fig 4. Integrated process to produce value-added phenolics and polymers from lignin in a biorefinery concept.

The polymorphic forms of vanillin were found to be dependent on the crystallization method as well as the temperature variation during the process. The slow evaporation method and cooling crystallization gave origin to the stable polymorph Form I, with higher yields than swift cooling and evaporative process with heat, that favored the nucleation of vanillin polymorphic Form II. Moreover, from evaporative processes, the vanillin crystals present a purity of about 95 %, while for cooling processes the purity is slightly higher, with values around 99 %. The different evaporative and cooling crystallization processes studied in this work proved to be valuable considering the attaining of vanillin crystals as the last step of the Integrated Process developed.

### Future Perspectives

Future work related to the stages of fractionation and purification of the added-value compounds (vanillin and syringaldehyde) should be included:

- optimization of membrane filtration conditions to find the best combination of operating temperature, feed flowrate, and transmembrane pressure that minimize fouling formation with improved membrane productivities;
- improvement of the recovery step of the phenolic aromatic products by adsorption/desorption cycles, including the study of adsorption conditions;
- the use of oxidized lignin fraction(s) with higher molecular weight (retentate in ultrafiltration process) in a synthesis route for lignin-based products, allowing the complete valorization of lignin;
- further research of the stage of crystallization processes, especially for hardwood lignins, that represents a huge challenge due to the presence of higher contents of syringaldehyde.

The results achieved and the upcoming work will allow the development of an efficient and environmentally friendly fractionation process, that represents the most important task towards the industrial application of lignin-derived aromatics.

### Related Sustainable Development Goals



### Outputs

#### PhD Theses

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- [2] Filipa Casimiro, Lignin from pulp mill sulfite liquors: Production of vanillin and syringaldehyde by an integrated process within the biorefinery concept, PDEQB, FEUP, 2023

#### Selected Publications

- [1] I. Mota et al., Separation and Purification Technology 197, 360 (2018)
- [2] E. Gomes et al., Separation and Purification Technology 216, 92 (2019)
- [3] I.F.Mota et al., Separation and Purification Technology 217, 108 (2019)
- [4] E. Gomes et al., Separation and Purification Technology 216, 92 (2019)
- [4] E. Gomes et al., Separation and Purification Technology 247, 116977 (2020)

#### Team

**Alírio Rodrigues**, Professor/Group Leader/Principal Researcher; Carina Costa, PostDoc Researcher; Inês Mota, PostDoc Researcher; **Elson Gomes**, PhD Student; **Filipa Casimiro**, PhD Student

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