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Lignin Oligomers from Mild Base-catalyzed Depolymerization for Potential Application in Aqueous Soy Adhesive as Phenolic Blends

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Abstract:

We report, for the first time, lignin oligomers prepared with base-catalyzed depolymerization have a great potential to be used as phenolic blends for enhancing the tensile shear strength of the soy protein adhesive. In this work, kraft lignin was used as a model for lignin waste feedstock for the preparation of lignin oligomers. The resulting lignin oligomers were directly used as the phenolic blend to soy protein adhesive. The tensile shear strength of the resulting soy protein-lignin oligomer adhesive was tested on plywood samples. The results showed that 50 wt.% loading (by solid content) of lignin oligomers prepared with base catalyzed depolymerized process at 170 °C has enhanced the bonding strength by 13.2%. It indicated that those oligomers were highly promising to be used as phenolic blends for the soy protein adhesive system. The lignin oligomers obtained the same process under different temperatures: 140 and 200 °C, were also tested for shear strength but showed lower bonding strength. In addition, the lignin oligomer blended adhesive was characterized for single lap shear strength after water-soaking process, Fourier transform infrared spectroscopy, simulated curing process in thermogravimetric analyzer, and thermal stability. The work showed that the base-catalyzed depolymerization with a mild reaction temperature at 170 °C could produce lignin oligomers with the best suitable molecule size, for crosslinking with the soy protein molecules, and result in improved shear strength of soy protein adhesive. The resultant soy protein-lignin adhesive had a decent dry shear strength of 1.46 MPa and wet shear strength of 0.62 MPa. The FTIR characterizations of lignin before and after treatment showed that the C-O intensity has changed significantly during the depolymerization process, indicating the cleavage of the β-O-4 bond in the raw lignin structure. Most importantly, the bio-adhesive is aldehyde-free and water-based, which makes this adhesive highly promising in interior applications and wood products industry.

Keywords: Lignin Oligomers; Depolymerization; Soy Adhesive; Aldehyde-free

1. Introduction

Soy protein adhesive has always been an interesting glue candidate for many adhesion industries because it is sustainable, low cost, and formaldehyde-free [1]. It was widely used to glue wood products industrially from 1930-1960 [2]. However, the soy protein adhesive has some drawbacks. It suffered from low water-resistance and relatively low bonding strength. As the adhesive technology developed, it was gradually replaced by formaldehyde-based adhesives such as phenol-formaldehyde and urea-formaldehyde adhesives. However, the formaldehyde emission has caused environmental and health concerns because of its possibility of causing cancer [3]. Recently, soy protein adhesive was revived because the adhesive system did not contain any formaldehyde. Thus, how to overcome the disadvantages such as relatively low bonding strength and poor water resistance has become the focus of the research in soy-based adhesive systems. In addition, it was still debatable that soy protein should be used for adhesive industry rather than only for food purposes.

Lignin is widely considered as a waste from the paper industry, usually in the form of kraft lignin. The kraft lignin was commonly burned as fuel in paper mills. The resulted carbonates can later be recovered in a lime kiln to regenerate cyclable alkali that will be reused in the kraft process [4]. Thus, taking good advantage of kraft lignin and creating value-added products from waste lignin is of great importance. The reactivity of kraft lignin is relatively low due to original molecular bone structure formed by random polymerization of three phenyl-propanol remained even after the pulping process. In addition, the molecular weight of kraft lignin is typically over 4000 Da, making it difficult to react with other substances. Therefore, certain chemical or biological treatments must be applied to make lignin more reactive before further being utilized. One common way is to reduce the molecule size while increasing the surface hydroxyl group. The building blocks of the lignin are p-hydroxyphenyl (H), guaiacyl (G), and syringyl (S), which contains many hydroxyl groups, both phenolic and alcohol type. These units initially crosslink with each other to form lignin structure. Thus, depolymerization of lignin could produce monomers and oligomers of or derived from these three basic units. It could be achieved by degrading lignin chemically with base-catalyzed depolymerization (BCD) process at an elevated temperature under a pressurized condition [5–7]. In addition, alkali was reported to activate the wood surface to improve the bonding

strength of between adhesive and wood [8]. Alkali could also improve the solubility of the lignin when alkaline water was used as a solvent [9,10].

General requirements for an acceptable soy protein adhesive are good water-resistance, low curing temperature, and proper viscosity. There are many previous efforts in crosslinking soy protein with lignin. Sorghum lignin has been downsized with extrusion process, leading to smaller particles, which was used for blending soy protein adhesive and improving its water resistance, both in neutral and basic conditions, as well as the shear strength [11]. Soy protein and lignin were electro-spined in the presence of poly (ethylene glycol) to manufacture sub-microfibers, during which the authors have pointed out that a higher lignin loading could result in an increased hydrogen bonding and a decreased secondary structure of soy protein [12]. There was also research that focused on using partial depolymerized lignin to prepare lignin amine, which was used to modify the soy protein adhesive and greatly improved the water resistance [13]. The interaction of soy protein and lignin was studied for the adhesion and showed that two key factors were molecule contact and surface energies [1]. One study reported the interaction of 11S and 7S structures in soy protein with cellulose and lignin, indicating that the protein favors the lignin surface over cellulose [14]. The interaction between the amino acids and lignin was via forming a quinone methide intermediate when coniferyl alcohol were tested for cross-coupling with three different types of amino acids [15]. Crosslinking agents were reported to modify the soy protein adhesives and improve the bonding strength, such as glycidyl methacrylate and polyethylene glycol diacrylate [16]. These agents also helped with the curing process. The curing of the soy protein adhesive was studied and suggested with a range of 120-170 °C [17,18]. However, these agents are typically very expensive and currently still under development.

To the best knowledge of the authors, study of soy protein crosslinking with lignin oligomeric fragments from mild temperature base-catalyzed depolymerization (140-200 °C) has not been previously reported. In addition, high loading (~50 wt%) of these lignin fragments in soy protein adhesive has seldom been tested for bonding strength previously. These lignin fragments are mainly hydrophilic oligomeric molecules. Just like soy protein molecules, these oligomeric fragments became much more soluble in alkaline solutions. This work has synthesized and characterized a bio-adhesive from soy protein and these lignin fragments with decent bonding strength and water resistance. Water was used as the only solvent. No aldehyde was involved in

the crosslinking reactions. The resulting adhesive was investigated in bonding strength, water resistance, surface functional groups, curing process, and thermal stability. This work will help better understand the interaction of lignin oligomers and soy protein in alkaline adhesive system, in which lignin oligomers and soy protein are much more soluble and easier to be dispersed and chemically tangled to each other.

2. Material and Methods

2.1. Preparation of Soy Protein Isolate Adhesive

Soy protein has been prepared by isolating it from defatted soy flour (Cargill Inc., USA). The soy flour contains 51 wt.% soy protein. Typically, 4 g of soy flour was dissolved in 50 ml deionized water. The pH of the soy flour solution was adjusted to 10.0 by NaOH. The solution was stirred for 1 hour. Soy protein from the flour was soluble at this pH, along with other contents from the flour which were soluble. Then, the solution was centrifuged at 5000 rpm for 8 min to separate the clear top and insoluble substance. The clear top was collected, acidified with HCl to adjust pH to 4.0, stirred for 1 hour, then centrifuged. The soy protein isolate was not soluble at pH value of 4.0. Thus, the insoluble white paste-like substance from the bottom of the centrifuge vial was soy protein isolate (SPI). The SPI was collected and dissolved in 50 ml deionized water that adjusted to pH 10, resulting in SPI solution for further preparation of soy-lignin adhesive. SPI solution itself was a relatively diluted pure soy protein isolate adhesive. Figure 1 has displayed the preparation of the SPI solution. The SPI adhesive in this work was prepared by evaporating the excessive water until the volume was 25% of the initial SPI solution.

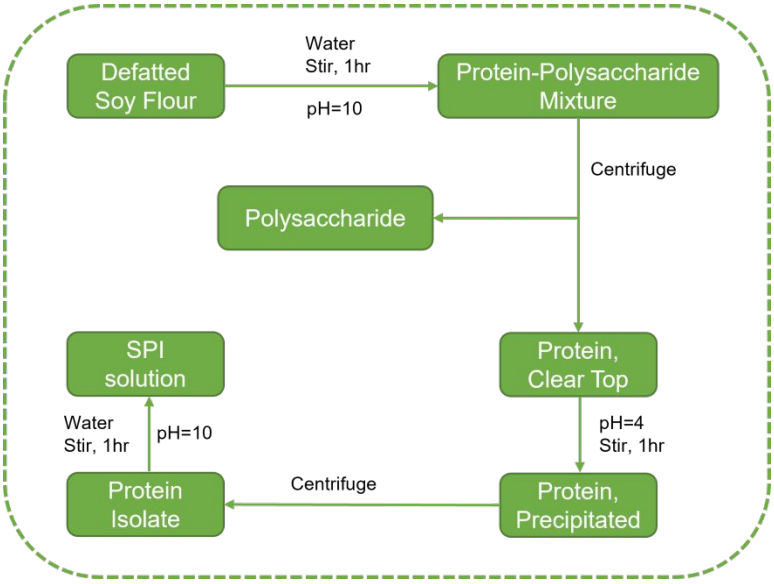

Figure 1. The isolation of soy protein isolate (SPI) and the preparation of SPI adhesive.

2.2. Base-catalyzed Depolymerization of Lignin to Oligomers

The partially degraded lignin (D-lignin or BCD Lignin) was prepared by base-catalyzed depolymerization of kraft lignin in pressurized autoclave. Typically, 2.5 g of kraft lignin (Sigma Aldrich, Product Number 370959) was dissolved in 50 ml deionized water. 0.185 g NaOH were added into the lignin solution to adjust the pH above 12. The NaOH weight percentage was 7.4 wt. % (based on lignin weight). The mixture was stirred for 1 hour. Then, the lignin solutions were transferred into the autoclave and held at 140, 170, and 200 °C. The pressure inside the autoclave was ranging from 100-150 psi. The residence time was 3 hours. The lignin solution after the treatment was used directly as the base-catalyzed depolymerized lignin oligomers. It was previously reported that the products were mainly lignin oligomers with negligible number of monomers (~3% or less) [19]. Thus, separation of monomers was not carried out before the preparation of bio-adhesive. In addition, the autoclave was sealed with air. The pressure during the depolymerization process was measured as 100~150 psi.

2.3. Preparation of Bio-adhesives

The SPI-D-lignin bio-adhesive was prepared by mixing SPI and D-lignin solutions with volume ratio of 2:1. The mixture was stirring for 1 hour. The adhesive solution was heated on a hotplate while stirring to remove excessive water. The temperature of the adhesive solution was measured

as 55 °C. It was reported that the soy protein and lignin undergone a pre-coupling reaction [20]. The adhesive solution was stirred during the entire process, which helped remove the excessive water. The resulting volume of the adhesive solution has been reduced to 25% of the initial value. A certain amount of water has been left on purpose to keep the viscosity at a proper level to maintain the flowability.

2.4. Solid Contents Measurement

The solid contents of three initial reactants have been measured. 2 ml liquid samples of SPI solution, untreated lignin solution, D-lignin solution have been dried at 105 °C for 24 hours. Untreated lignin solution was prepared with 50 ml water, 0.185 g NaOH, and 2.5 Kraft Lignin (same formation with D-lignin solution before treatment). 1 ml liquid samples of final soy protein-lignin adhesives were dried using the same condition for: reactants SPI solution and lignin solution, SPI-lignin adhesive, SPI-D-lignin adhesive. Since the adhesives were prepared by removing excessive water, the solid contents mass ratio was controlled to 1:1 for SPI and D-lignin in the eventual adhesive samples. The solid contents of the adhesive and related components have been measured and listed in Table 1.

Table 1. Solid contents of SPI, Lignin and Bio-adhesives in this study

Samples	Solid Content (mg/ml)	Density (g/ml)
Soy Protein Isolate (SPI) solution	13.01	1.00
Lignin, untreated (Lignin)	23.69	1.02
Lignin, degraded (D-lignin, 170 °C)	23.77	1.03
Adhesive, SPI and untreated lignin	138.19	0.99
Adhesive, SPI and D-lignin, 170 °C	113.08	1.02

2.5. Characterization of Surface Functional Groups

The functional groups of dried solid samples for SPI, untreated lignin, D-lignin, and SPI-D-lignin adhesive were characterized using Fourier Transform Infrared (FTIR) spectroscopy. The purpose is to study the change of the functional groups during the depolymerization and the crosslinking reactions. The samples for FTIR were dried at 60 °C in vacuum oven for 48 hours. The samples were dried at relatively low temperatures to preserve the functional groups on these molecules.

2.6. Single Lap Shear Strength

The plywood samples were cut into required dimensions as 100 mm long, 25 mm wide, 2.5 mm thick. The test was carried out based on ASTM D906. The plywood samples were applied with 125 µl of adhesive aqueous solution on a 25 by 25 mm area. The samples were cold pressed for 2 mins, then hot pressed with 0.0235 MPa inside the 120 °C oven to simulate the hot press scenario. The hot presser was not used in this work because the dimensions of the plywood samples were changing significantly inside the hot presser, especially the thickness of the wood samples. The wood samples applied with adhesives were cured in 30 mins. Every adhesive was tested with 5 repetitions of the glued plywood samples. The wet strength was collected with bonded plywood samples soaked in water at room temperature for 3 hours. In addition, a commercial soy protein adhesive, BreezeBond™, from Specialty Organics (California, USA) was tested using the same method and compared.

2.7. Curing Process Simulation

The curing process was simulated in the thermogravimetric analyzer (TG), which has an in-situ differential scanning calorimetry (DSC) that can monitor the heat flow. The curing simulation was carried out with different mass loading of SPI-D-lignin (BCD lignin at 170 °C) adhesive while the applied area of adhesive remains unchanged. The curing temperatures were simulated at 120 °C. The curing ramping rates were selected as 5 and 10 °C/min. The curing process was carried out with air flow.

2.8. Adhesive Thermal Stability

The thermal stability of the adhesive was tested using TG. The characterization was carried out with a 10 °C/min ramping rate. The degradation temperatures were selected as 250 °C. The test was carried out in nitrogen flow. The adhesive solids were pre-dried at 60 °C in a vacuum oven before being tested for thermal stability.

2.9. ^{1}H NMR Spectra Analysis

0.7 ml D_2O solutions of lignin oligomer, soy protein, and the as-prepared bio-adhesives before and after curing process were characterized with Nuclear Magnetic Resonance (NMR) spectroscopy. The solution samples were loaded into a 5 mm tube for NMR measurement. The ^{1}H NMR spectra were collected on a 600 MHz NMR spectrometer (Varian, USA). The ^{1}H NMR spectra of the residual proton peaks (HDO) of D_2O was referenced at 4.75 ppm. Sodium

trimethylsilylpropanesulfonate (DSS) was added into each sample as an internal reference during the preparation of D_2O solutions of samples. The ^1H NMR spectrum of DSS consisting of chemical shifts at 2.91 (CH_2), 1.75 (CH_2), 0.63 (CH_2), and 0 ppm ($Si(CH_3)_3$).

3. Results and Discussion

3.1. Yield and Molecular Weight of Lignin Oligomers

Bernhardt et al.[19] has conducted hydrothermal base catalyzed depolymerization of pine wood based kraft lignin in pilot scale. They have reported BCD experiments in two volumes of reactors, 1 L and 0.055L. The volume of the BCD of the current work was extremely close to the 0.055 L reported. Thus, we are referring to their 0.055 L reactor for the approximate yield of our experiment. Their work has found that, with 1.67 wt.% NaOH, the oligomer yield was an average of 84.0 wt.% at 240 °C, 79.1 wt.% at 270 °C, and 70.4 wt.% at 300 °C. While with 2.5 wt.% NaOH, the oligomer yield was an average of 81.8 wt.% at 240 °C, 76.8 wt.% at 270 °C, and 71.4 wt.% at 300 °C. Thus, it was reasonable to believe that our yield should be estimated at 85% or higher. It was worth noting that the separation of oligomer and unreacted lignin was extremely different at lab scale. Thus, the current work must take an estimation of the actual oligomer yield. The acid precipitation method can only separate the phenolic products with light molecular weight (<1000 Da). Oligomer and unreacted lignin always precipitated together during the acid precipitation.

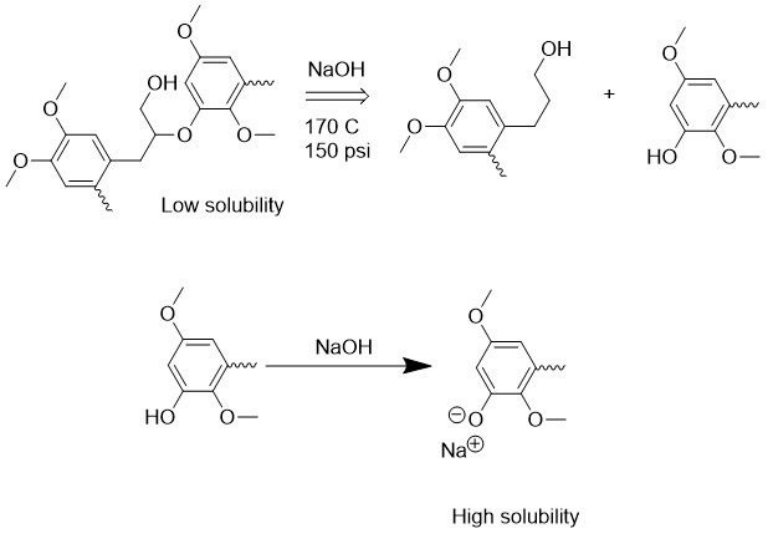

Figure 2. Cleavage of β-O-4 bond results in smaller and more water-soluble lignin oligomers.

The molecular weight of lignin oligomers prepared by base-catalyzed depolymerization were widely reported. It was typically characterized by gel permeable chromatography (GPC). Due to equipment limitations within our institution, we were not able to characterize the lignin oligomers using a GPC. Thus, we are presenting a reasonable estimation of the molecular weight range for our lignin oligomers prepared in the lab, based on the original molecular weight on display of the starting kraft lignin purchased. In addition, at higher alkali concentration, lignin molecular weight, in general, was less dependent on temperature [21]. Therefore, based on the previously reported work in Table 2, the molecular weight of our lignin oligomer was reasonably estimated to be within the range of 1000-4000 Da.

The base-catalyzed depolymerization not only reduced the size of the lignin molecule, but also increased the water-solubility of the lignin molecule. The phenolic hydroxyl could easily react with NaOH and form sodium phenoxide, which was much more soluble than the original lignin molecular, shown in Figure 2.

Table 2. Molecular weight of lignin oligomer from base-catalyzed depolymerization reported in previous work.

Reference	Lignin Type	Treatment	**Alkali**	**Oligomer MW**
Rodriguez **et al. 2020** [21]	Hydrolysis	60-240 °C, 0.7-20 bar, 40 min	1% and 2% w/v, NaOH	>1000 Da
Erdocia et al. 2014 [22]	Organosolv	300 °C, 80 min, 9 MPa	4 wt.% NaOH w/w	2000-4500 Da
Katahira et al. 2016 [5]	Hydrolysis, Kraft	270-330 °C, 40 min	2 wt. %, 4 wt.% NaOH	5000 Da
Bernhardt et al. 2021 [19]	Kraft	240-300 °C, 250 bar	1.67 wt.% or 2.5 wt.% NaOH	1000-3000 Da

3.2. Single Lap Shear Strength

The soy protein-based adhesives with 50% loading of lignin based on mass were crosslinked with partially depolymerized lignin treated at three different temperatures. The kraft lignin was treated with base-catalyzed depolymerization which was slightly modified. The process was referred to a previous work completed by National Renewable Energy Laboratory [5], which has a NaOH usage of 2 wt.% to 4 wt.% and at a higher treatment temperature of 270-330 °C. In the current work, the

temperature was reduced to the range of 140 to 200 °C to avoid the total decomposition of the intermediate lignin oligomers. These oligomers might crosslink with the soy protein isolate molecules, leading to a better bonding strength. All three SPI-Lignin adhesives were characterized for dry single lap shear strength.

In Figure 3, it showed that the bonding strength decreased from 1.29 MPa to 1.03 MPa when 50% of untreated lignin was added into the soy protein adhesive. When it came to the incorporation of depolymerized lignin, the results showed that adding lignin treated at 170 °C at a 50% mass loading had a highest 1.46 MPa, which was higher than 1.09 MPa and 0.85 MPa for 140 and 200 °C, respectively. It was worth noting that only the one with 170 °C treated lignin had a strength value surpassed that of the SPI adhesive, which indicated that the partially depolymerized lignin under this condition could be used to enhance the bonding strength of the soy protein isolate adhesive. The improved bonding strength could be due to the oligomers and monomers generated during the depolymerization process at this temperature having a better crosslinking network with the molecules of the soy protein isolate. In addition, BreezeBond™, a commercial soy protein, has been evaluated using the same testing method. The result showed that the commercial soy protein has an expected higher single lap shear strength of 2.07 MPa. Thus, the D-lignin at 170 °C and the resultant adhesive was selected for further characterizations later in this work. The resistance to water was tested with plywood samples bonded with the SPI-Lignin 170 °C. The bonded samples were soaked in water at room temperature for 3 hours. As shown in Figure 4, the bonding strength has decreased from 1.46 MPa to 0.62 MPa. The adhesive system of SPI-Lignin 170 °C has been selected for more characterizations in the following sections. A lignin-based resin was adopted to improve the properties of a soy flour-based adhesive, in which a dry bonding strength of 1.32 MPa and a wet bonding strength of 0.69 MPa were achieved in the absence of external crosslinking agent [23].

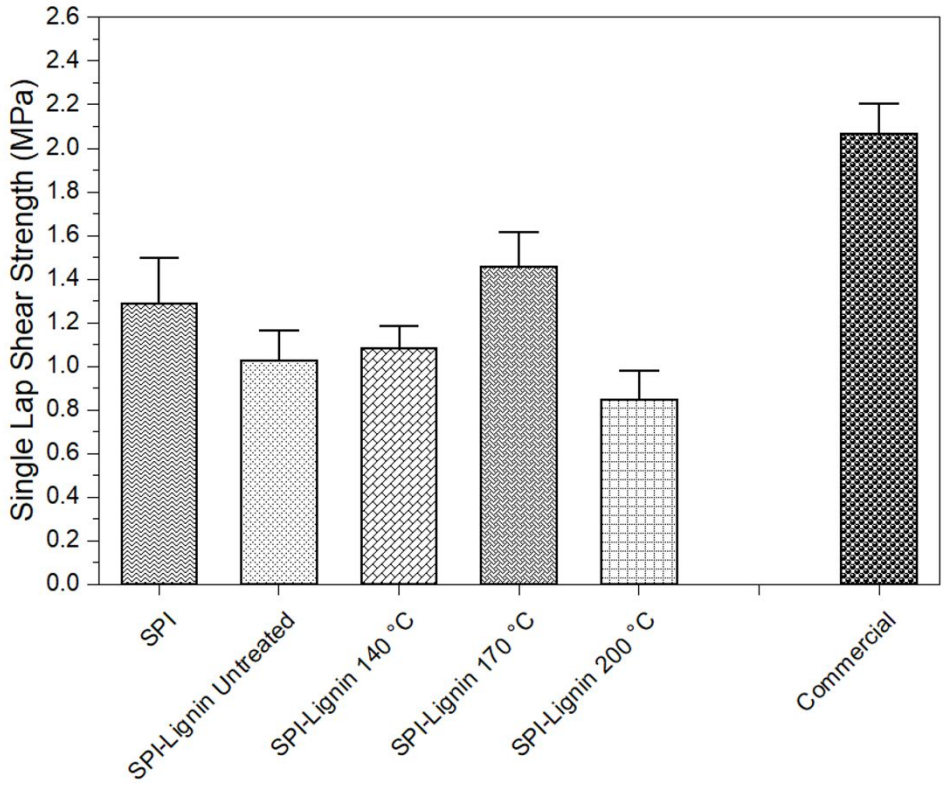

Figure 3. Bonding strength of bio-adhesives as prepared in this work with comparison to the commercial soy protein adhesive (BreezeBond™, Specialty Organics, California, USA)

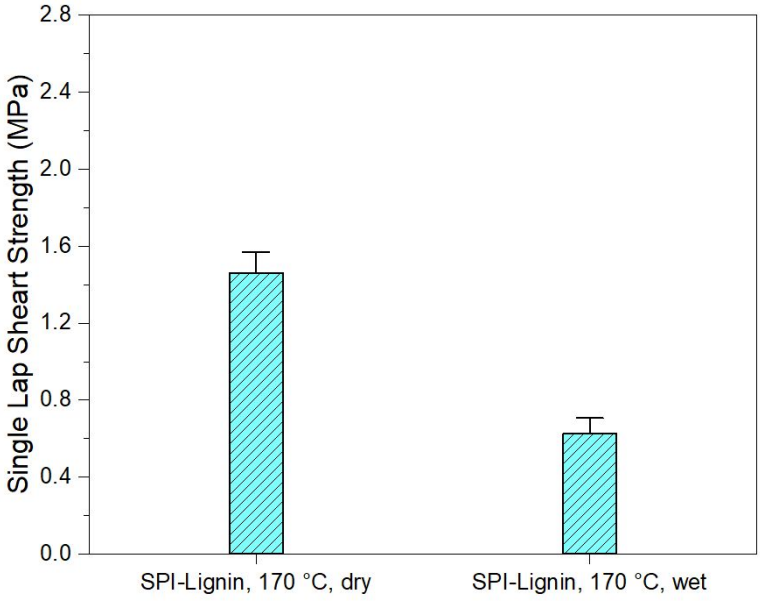

Figure 4. Water resistance of SPI-Lignin 170 °C regarding to bonding strength

3.3. Chemical Analysis of Depolymerized Lignin

The base-catalyzed depolymerization was used to break lignin into fragments. FTIR analytical approach was applied to study the surface functional groups of the partially degraded lignin and the untreated lignin. The assignment of the transmittance peaks was listed in Table 2, which was created based on a previous report [24]. Figure 5a and 5b has shown the comparison of peaks between untreated lignin and the partially degraded lignin under base-catalyzed depolymerized (BCD) process, which were used to prepare the adhesives. The treated lignin has shown no visible peak shifting. It indicated that the lignin molecule frame was deconstructed to mainly lignin oligomers, rather than lignin derived monomers. It was reported previously that lignin from BCD process at 270 °C treated with 4% NaOH had resulted in low molecular weight (M_w) chemical species in 200~350 Da range and high M_w species in 350~2000 Da range, which were denoted as derivative monomers and oligomers, respectively [5]. In the current work, the treatment temperature was much lower than 270 °C. Since there was no visible change of peak positions on the FTIR spectroscopy, it was reasonable to believe that the lignin fragments were mainly lignin oligomers. The SPI-lignin adhesive using untreated and BCD lignin at 170 °C has been shown in Figure 6. The peak positions and intensity did not show visible changes. Two different lignin might form crosslinking structures with soy protein molecules, resulting in different strength. However, the functional groups formed could be similar. Thus, FTIR was expected to show little difference when it came to final adhesives products.

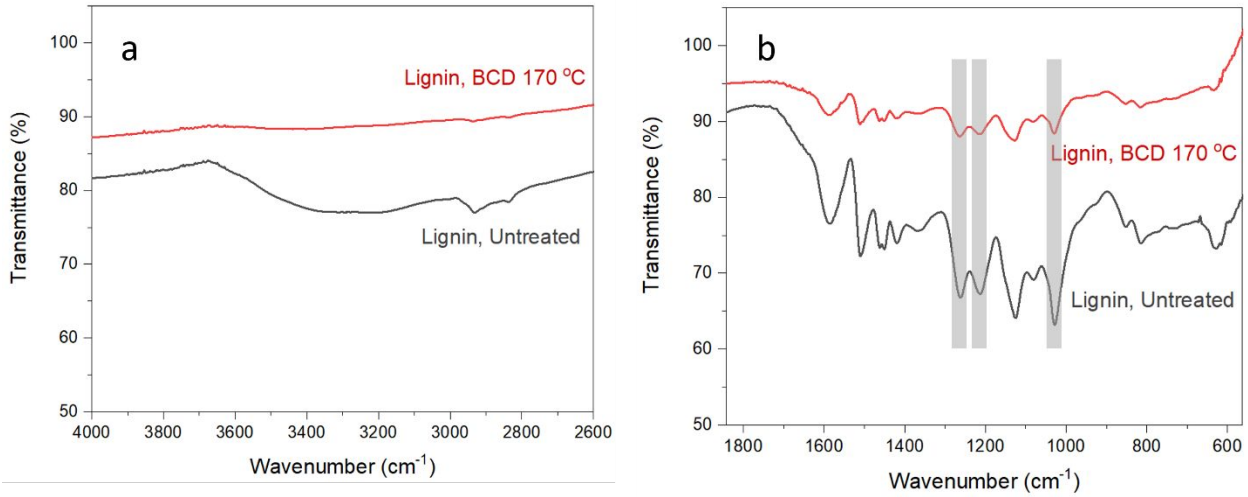

Figure 5. FTIR of lignin treated with BCD and untreated lignin: (a) transmittance from 2600 to 4000 cm^{-1} and ratio; (b) transmittance of fingerprint region and ratio.

Table 3. FTIR peak assignments for BCD and untreated lignin

Wavenumber (cm^{-1})	Assignments of Chemical Functional Groups
3400	O-H stretching, phenolic & aliphatic
2930	C-H stretching, $-CH_3$ & $-CH_2$
2837	C-H stretching, $-O-CH_3$
1586	C-C stretching, aromatic
1508	C-C stretching, aromatic
1455	C-H deformation, asymmetric, $-CH_3$ & $-CH_2$
1419	C-C stretching, aromatic, with C-H in-plane deformation
1261	C-O stretching vibration of secondary alcohol
1214	C-O(H) & C-O, phenolic OH & ether in syringyl and guaiacyl
1125	Aromatic -CH in plane deformation, syringyl
1028	C-O(H) & C-O(C), first order aliphatic -OH & ether
813	C-H out of plane, aromatic

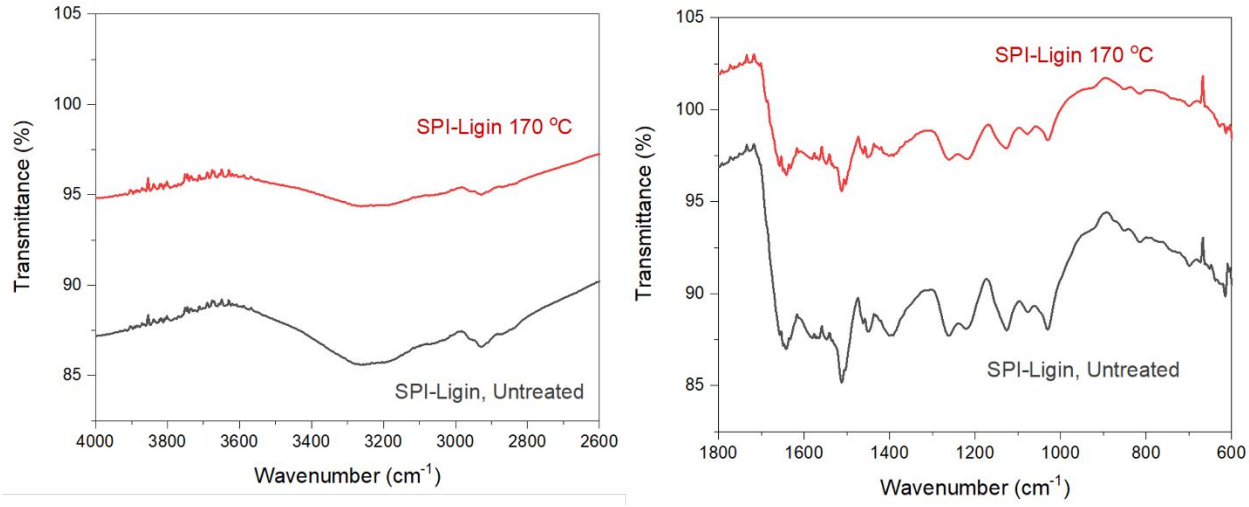

Figure 6. FTIR of SPI-Lignin 170°C (D-Lignin, 50 wt.%) and SPI-Untreated Lignin

In the fingerprint area (600-1800 cm^{-1}), it was worth noting that 1028, 1214, and 1261 cm^{-1} had extremely visible variations of intensity before and after BCD treatment. According to Table 3, these are all related to the C-O and C-OH bond inside the lignin molecules. It was evidenced that

cleavage of β-O-4 ether, as shown in Figure 2 was the major reaction for BCD process. The BCD process could reduce the molecular weight of lignin and increase its reactivity [25].

3.4. Interaction of Lignin Oligomer and Soy Protein

Figure 7 showed the ^{1}H NMR spectra obtained at +25 °C for 10 mg of each following sample in 0.7 ml D_2O: (A) crystal of lignin oligomer, (B) crystal of soy protein, (C) crystal of lignin oligomer (50 wt.%) and soy protein (50 wt.%) mixture at 25 °C, and (D) crystal of lignin oligomer (50 wt.%) and soy protein (50 wt.%) mixture after curing process (120 °C for 30 min). The internal reference peaks of DSS were highlighted. To have better comparison, Figure 8 displayed the amplified spectral regions of the ^{1}H NMR spectrum for each sample. The individual lignin oligomer (Sample A) and soy protein (Sample B) spectra displayed the same HDO chemical shift. However, the mixture of lignin oligomer and soy protein mixture at 25 °C (Sample C) showed a downfield shift (Figure 8a), indicating Sample C had interactions with water that was obviously different from either lignin oligomer/water or soy protein/water interactions. This was direct evidence that there were interactions between lignin oligomer and soy protein when they were mixed in the presence of water. As shown further in Figure 8b and 8c, we have found upfield shift (3.20~3.08 ppm) and downfield shift (2.68-2.62 ppm) due to interactions between soy protein and lignin oligomer [26,27]. The Sample D has shown no such interaction because the curing process at 120 °C for 30 min had erased this interaction. This observation indicated that there were indeed interactions between soy protein and lignin oligomer when mixed to prepare bio-adhesive.

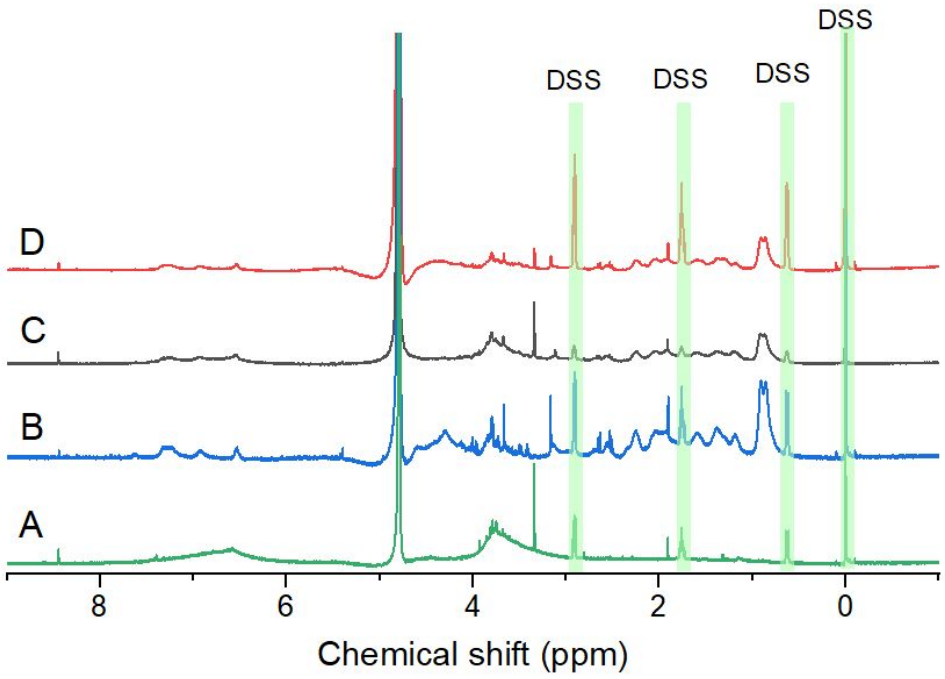

Figure 7. ^{1}H NMR spectra of lignin oligomer (A), soy protein (B), bio-adhesive (C), and adhesive after curing process (D)

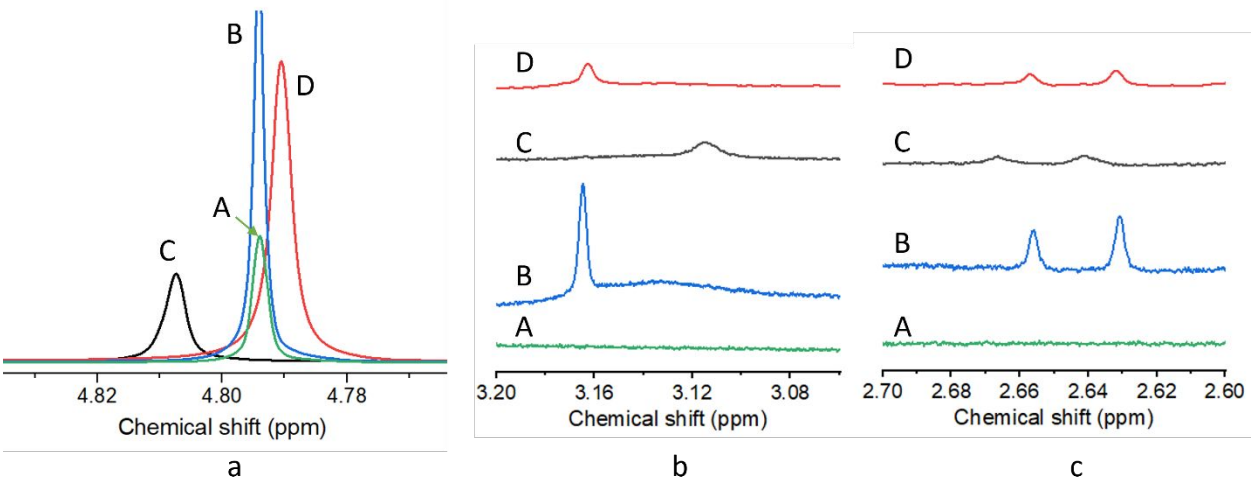

Figure 8. amplified spectral regions of the ^{1}H NMR spectrum for lignin oligomer (A), soy protein (B), bio-adhesive (C), and adhesive after curing process (D)

3.5. Possible Reactions in Lignin Depolymerization and Adhesive Crosslinking

Base-catalyzed depolymerization typically led to the cleavage of β-O-4 or α-O-4 ether bonds in the lignin structures. According to previous literature [28,29], possible lignin oligomeric structures have been listed in Figure 9. Kraft lignin from base-catalyzed depolymerization in NaOH at temperatures from 170 to 250 °C has shown that the molecular weight of the lignin fragments decreased when the treatment temperature increased [30]. These lignin oligomers generally had higher reactivity than kraft lignin due to higher content of hydroxyl group. Due to the OH^- anion in the resulted solution, the dissociation of phenolic and alcohol H^+ became more active, which could improve the solubility of these lignin fragments in water. During the curing process, these fragments were crosslinked with soy protein molecules in alkaline solution (pH~10). The crosslinking reactions took place at 120 °C for 30 min. Possible reactions during curing process have been listed in Figure 10. The phenolic and aliphatic OH from lignin oligomer reacted with hydroxyl, carboxylic and primary amine groups in soy protein molecules via condensation. The crosslinked molecular framework was also formed between the resultant adhesive and wood surface during the curing process. The hydroxyl groups from the wood surface went through condensation reactions with soy protein and lignin oligomers, forming strong chemical bonds between dried adhesive and wood.

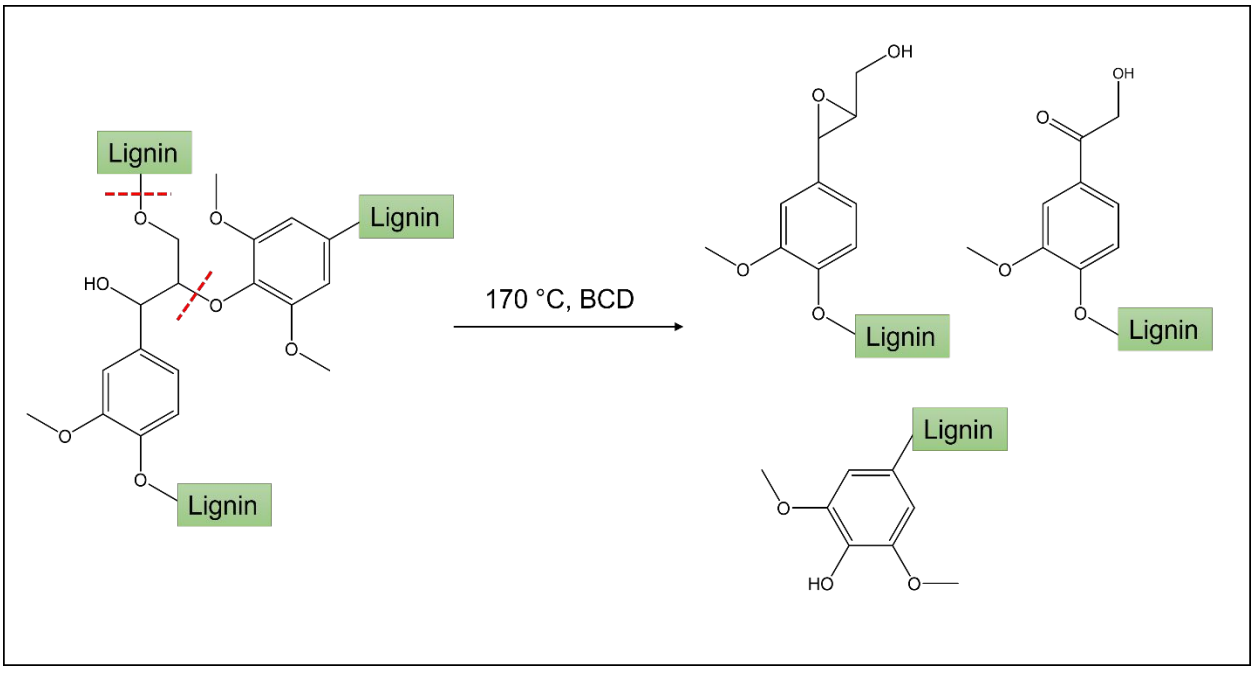

Figure 9. Cleavage of ether bonds and possible resultant lignin oligomers.

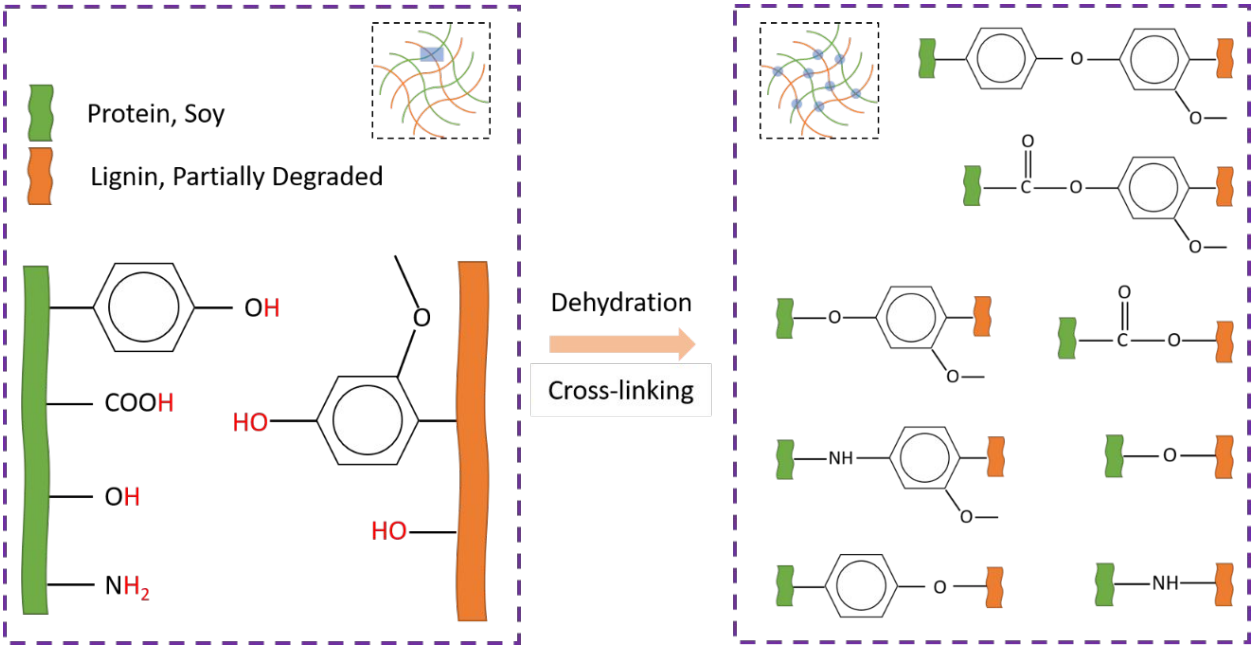

Figure 10. Possible crosslinking reactions between lignin fragments and soy protein molecules.

3.6. Curing of Adhesive

The curing of the soy protein-lignin adhesive has been simulated using a thermogravimetric analyzer (TG) and in-situ differential scanning calorimetry (DSC). Adhesive system SPI-Lignin 170 °C was selected for the characterization. The curing temperature was set as 120 °C. The curing temperature has been optimized before the curing simulation. Figure 11a shows the TG measurement of different adhesive loadings, while the applied adhesion area for these three loadings were identical. All adhesive loading was cured within 25 min. When the temperature ramping rate is 5 °C/min, higher adhesive loading required longer time to cure. The results between different temperature ramping rates have indicated that higher ramping rate led to a shorter curing time requirement. The curing process of soy protein modified with trypsin was investigated for wood bonding, indicating that the curing time should be kept within the range of 0.5 to 1.0 hour, over which the bonding strength would decrease [31]. The DSC results in Figure 11b have indicated that the curing of the selected adhesive was an overall endothermic reaction due to the heat required to evaporate the solvent (water) in the adhesive system. Possible reactions for crosslinking process were listed in Figure 10. Most of these are either dehydration reactions (forming ester or ether) or

secondary amine forming reactions. The soy protein-lignin was reported to have a higher endothermic heat requirement when compared to that of pure protein [32].

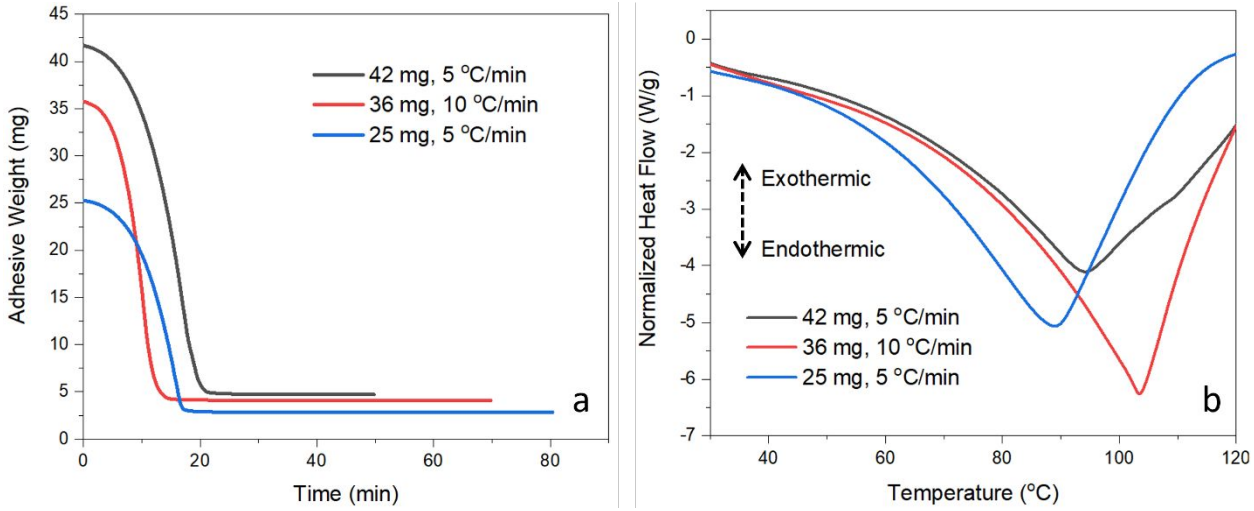

Figure 11. The thermogravimetric measurement (a) and differential scanning calorimetry (b) characterization of soy-protein adhesive curing process.

3.7. Thermal stability of adhesive system

The thermal stability of the components of the adhesive has been investigated with a temperature up to 250 °C. The ramping rate was set as 10 °C/min. All components were pre-dried with a vacuum oven at 60 °C for 48 hours. As shown in Figure 12, the depolymerized lignin has the highest thermal stability. The weight loss was less than 4% when the temperature increased from 50 to 250 °C. The SPI has the least thermal stability. The derivative TG analysis shown in Figure 12b has indicated that the SPI-Lignin 170 °C adhesive was mainly determined by the SPI, because of the consistent trends between thermal degradations of SPI and SPI-Lignin 170 °C adhesive. It indicated that the depolymerized lignin could act as a filler to improve the thermal stability of the SPI based adhesive. The crosslinking framework formed between the SPI and partially degraded lignin could also contribute to the thermal stability of the resultant adhesive. Alkali lignin was reported to be used for increasing the onset temperature of soy protein based films, leading to a more thermally stable structure [33]. Similar results were also found when lignosulfonate was adopted to improve the thermal stability of soy protein plastic [34], which indicated that the increased loading of lignosulfonate, a soluble lignin derivatives, resulted in a higher onset temperature.

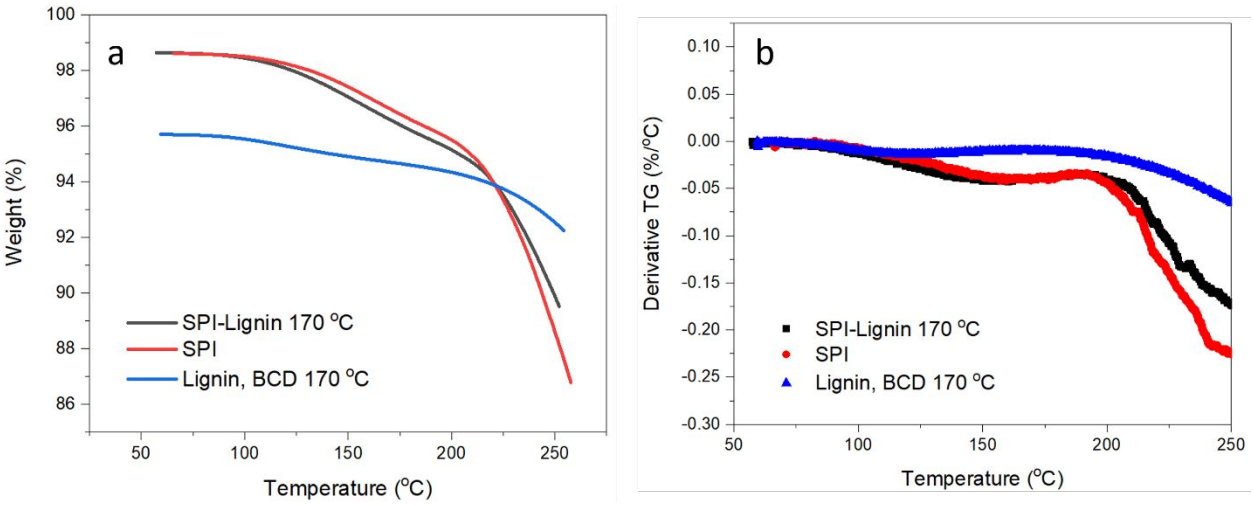

Figure 12. Thermal stability of SPI, BCD lignin, and bio-adhesives.

4. Conclusion

Aqueous aldehyde-free soy protein adhesives with lignin oligomers as phenolic blends were successfully prepared and enhanced bonding strength. Kraft lignin as a model lignin waste was partially depolymerized to lignin oligomers using mild base-catalyzed depolymerization at 170 °C. The lignin oligomers were used as phenolic blend for the soy protein adhesive. The results showed that 50 wt.% loading of lignin (based on solid content) has increased the bonding strength by 13.2%. When the temperature of BCD treatment was higher or lower than 170 °C, the bonding strength was lower than the control soy adhesive. It indicated that the smaller lignin fragments were not necessarily beneficial to the bonding strength. The results showed that when lignin treated with 170 °C was used as the crosslinking agent, the bonding strength surpassed the soy protein isolate adhesive (control). The FTIR results confirmed that the products of the BCD process were mainly lignin fragments in oligomeric range. The results also showed that partially degraded lignin could help improve the thermal stability of the soy protein-based adhesive. This encouraging result has indicated that lignin oligomers could be used to crosslink with soy protein isolate and form an aldehyde-free bio-adhesive that has decent bonding strength and water resistance. This work has demonstrated potentially the environmental, health and economic benefits of the bio-adhesive.

CRediT authorship contribution statement

Changle Jiang: conceptualization, data curation, formal analysis, investigation, methodology, writing-original draft, and writing-review and editing. **Jianli Hu:** conceptualization, data curation, formal analysis, investigation, methodology, writing-review and editing, project administration, funding acquisition, resources, and supervision. **Chao Zhang:** formal analysis, investigation, and methodology. **Gangarao Hota:** methodology. **Jingxin Wang:** project administration, funding acquisition. **Novruz Akhmedov**: formal analysis, investigation, and methodology.

Declaration of competing interest

The authors declare no competing financial interest.

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References

1 R. J. Ross, *USDA For. Serv. For. Prod. Lab. Gen. Tech. Rep. FPL-GTR-190, 2010 509 p. 1 v.*

2 Y. Liu and K. Li, *Macromol. Rapid Commun.*, 2002, **23**, 739–742.

3 M. Podlena, M. Böhm, D. Saloni, G. Velarde and C. Salas, *Polymers (Basel).*, 2021, **13**, 1–16.

4 H. Tran and E. K. Vakkilainnen, *Tappi Kraft Pulping Short Course*, 2008, 1–8.

5 R. Katahira, A. Mittal, K. McKinney, X. Chen, M. P. Tucker, D. K. Johnson and G. T.

Beckham, *ACS Sustain. Chem. Eng.*, 2016, **4**, 1474–1486.

6 R. Chaudhary and P. L. Dhepe, *Green Chem.*, 2017, **19**, 778–788.

7 M. Otromke, R. J. White and J. Sauer, *Carbon Resour. Convers.*, 2019, **2**, 59–71.

8 R. A. Young, M. Fujita and B. H. River, *Wood Sci. Technol.*, 1985, **19**, 363–381.

9 E. Melro, A. Filipe, D. Sousa, A. J. M. Valente, A. Romano, F. E. Antunes and B. Medronho, *Int. J. Biol. Macromol.*, 2020, **148**, 688–695.

10 S. Maitz, W. Schlemmer, M. A. Hobisch, J. Hobisch and M. Kienberger, *Adv. Sustain. Syst.*, , DOI:10.1002/adsu.202000052.

11 Z. Xiao, Y. Li, X. Wu, G. Qi, N. Li, K. Zhang, D. Wang and X. S. Sun, *Ind. Crops Prod.*, 2013, **50**, 501–509.

12 C. Salas, M. Ago, L. A. Lucia and O. J. Rojas, *React. Funct. Polym.*, 2014, **85**, 221–227.

13 J. Xin, P. Zhang, M. P. Wolcott, J. Zhang, W. C. Hiscox and X. Zhang, *J. Polym. Environ.*, 2017, **25**, 599–605.

14 C. Salas, O. J. Rojas, L. A. Lucia, M. A. Hubbe and J. Genzer, *ACS Appl. Mater. Interfaces*, 2013, **5**, 199–206.

15 F. Cong, B. G. Diehl, J. L. Hill, N. R. Brown and M. Tien, *Phytochemistry*, 2013, **96**, 449–456.

16 J. Luo, J. Luo, C. Yuan, W. Zhang, J. Li, Q. Gao and H. Chen, *RSC Adv.*, 2015, **5**, 100849–100855.

17 X. Li, Y. Li, Z. Zhong, D. Wang, J. A. Ratto, K. Sheng and X. S. Sun, *Bioresour. Technol.*, 2009, **100**, 3556–3562.

18 H. Yin, E. Zhang, Z. Zhu, L. Han, P. Zheng, H. Zeng and N. Chen, *ACS Appl. Polym. Mater.*, 2021, **3**, 1032–1041.

19 J. J. Bernhardt, B. Rößiger, T. Hahn and D. Pufky-Heinrich, *Ind. Crops Prod.*, , DOI:10.1016/j.indcrop.2020.113119.

20 C. Florencio, A. C. Badino and C. S. Farinas, *Bioresour. Technol.*, 2016, **221**, 172–180.

21 A. Rodriguez, D. Salvachúa, R. Katahira, B. A. Black, N. S. Cleveland, M. Reed, H. Smith, E. E. K. Baidoo, J. D. Keasling, B. A. Simmons, G. T. Beckham and J. M. Gladden, *ACS Sustain. Chem. Eng.*, 2017, **5**, 8171–8180.

22 X. Erdocia, R. Prado, M. Á. Corcuera and J. Labidi, *Biomass and Bioenergy*, 2014, **66**, 379–386.

23 X. Zhang, Y. Zhu, Y. Yu and J. Song, *Polymers (Basel).*, 2017, **9**, 261.

24 M. N. M. Ibrahim, N. Zakaria, C. S. Sipaut, O. Sulaiman and R. Hashim, *Carbohydr. Polym.*, 2011, **86**, 112–119.

25 H. Z. Chen, Z. Y. Li, X. Y. Liu, Y. M. Tian, L. Yang and Z. C. Wang, *J. Appl. Polym. Sci.*, , DOI:10.1002/app.42176.

26 F. Kong, K. Parhiala, S. Wang and P. Fatehi, *Eur. Polym. J.*, 2015, **67**, 335–345.

27 S. Mehra, S. Nisar, S. Chauhan, V. Singh and S. Rattan, *ACS Omega*, 2020, **5**, 21610–21622.

28 T. J. McDonough, *TAPPI J.*, 1993, **76**, 186–193.

29 N. Abad-Fernández, E. Pérez and M. J. Cocero, *Green Chem.*, 2019, **21**, 1351–1360.

30 O. Y. Abdelaziz, K. Li, P. Tunå and C. P. Hulteberg, *Biomass Convers. Biorefinery*, 2018, **8**, 455–470.

31 U. Kalapathy, N. S. Hettiarachchy, D. Myers and M. A. Hanna, *J. Am. Oil Chem. Soc.*, 1995, **72**, 507–510.

32 S. Pradyawong, G. Qi, N. Li, X. S. Sun and D. Wang, *Int. J. Adhes. Adhes.*, 2017, **75**, 66–73.

33 E. M. Zadeh, S. F. O'Keefe and Y. T. Kim, *ACS Omega*, 2018, **3**, 7388–7398.

34 J. Huang, L. Zhang and F. Chen, *J. Appl. Polym. Sci.*, 2003, **88**, 3284–3290.