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Mika Henrikki Sipponen,* Orlando J. Rojas, Ville Pihlajaniemi, Kalle Lintinen, Monika Österberg

*Corresponding author E-mail: mika.sipponen@aalto.fi

Calcium chelation of lignin from pulping spent liquor for water-resistant slow-release urea fertilizer systems

Mika Henrikki Sipponen,^{,1,2} Orlando J. Rojas,² Ville Pihlajaniemi,^{1,3} Kalle Lintinen,¹ Monika Österberg²*

*Corresponding author E-mail: mika.sipponen@aalto.fi

¹Aalto University School of Chemical Technology, Department of Biotechnology and Chemical Technology, Espoo, Finland
Kemistintie 1, 02150 Espoo, Finland (street address); P.O. Box 16300 FI-00076 Aalto (mailing address)

²Aalto University School of Chemical Technology, Department of Forest Products Technology, Espoo, Finland
Vuorimiehentie 1, 02150 Espoo, Finland (street address); P.O. Box 16100 FI-00076 Aalto (mailing address)

³VTT Technical Research Centre of Finland Ltd, Espoo, Finland
Tietotie 2, 02150 Espoo, Finland (street address); P.O. Box 1000, FI-02044 VTT, Finland (mailing address)

ABSTRACT

Slow-release fertilizers represent a possible large-scale application for plant polymers. Here we show a facile way to stabilize urea in fertilizer systems by lignin. Chelation of kraft black liquor with calcium acetate at pH 13 precipitated lignin as a calcium complex (Ca-lignin), which offered beneficial effects if compared to those from lignin obtained by precipitation at low pH (Acid-lignin). The reduced affinity of water to Ca-lignin was exploited in the formulation of slow release fertilizers comprising wheat straw sections impregnated with Ca-lignin in molten urea. Compared to the case of Acid-lignin, immersion in water was slowed down more extensively by Ca-lignin. After 24 h incubation at low moisture conditions, the highest proportion of urea retained in the Ca-lignin/straw fertilizer system was 58%. The water resistance of Ca-lignin was explained by a lower aqueous solubility that differed from the typical pH-dependent solubility of Acid-lignin. Electron microscopy, infrared spectroscopy, and accessible surface areas suggested that Ca-lignin consisted of less densely packed molecules organized as calcium-chelated chains. Overall, the controlled water-solubility of lignin precipitated by metal cations is greatly beneficial in fertilizer systems and can open new opportunities in material development (permeable films and others).

INTRODUCTION

More efficient use of fertilizers is needed to sustainably increase agricultural productivity in response to population growth. Utilization of waste streams for fertilizer production is important for advancing sustainability of industry and transition to circular economy. Recently, wastewaters from metal and dairy processing plants were used to produce zinc-containing chelated micronutrient fertilizers.¹ Production of nitrogen-based fertilizers consumes energy, for example in the cryogenic distillation of N₂ from air, and additionally, requires hydrogen derived from fossil carbon sources. Urea is one of the most widely used N-fertilizers today, but its microbial decomposition and uncontrolled leaching from soil are major hurdles that need attention.²

Many attempts have been made to stabilize highly water-soluble urea in fertilizers. For example, dissolution of urea has been retarded by encapsulation in polymers,³⁻⁵ by using resin or polymer coatings in urea granules,⁶⁻¹⁰ or by chemically reacting urea with compounds such as formaldehyde.^{11,12} Some drawbacks of these approaches include the inherent cost of chemical modifications, usage of harmful chemicals, and limited biodegradability of the polymeric additives. In contrast, renewable plant biomass could provide sustainable material for the preparation of slow release fertilizers.

Lignin is the main phenolic polymer of lignocellulose with well-suited features for slow release formulations of urea. Lignin is available in abundant quantities from the existing cellulose pulping processes, which makes it an economically attractive raw material. The most common technical lignin is kraft lignin, which is not fully soluble in the pH range typical of soils. Lignin serves as a precursor to humic substances that are important constituents of the soil organic carbon. Moreover, the abundant phenolic functionalities

of lignin could stabilize soil's organic nitrogen,¹³ and lignin or low molecular weight phenolic compounds released upon its microbial degradation, could inhibit two detrimental events occurring in soils: nitrification,^{14,15} and the unwanted volatilization due to soil urease activity.¹⁶

Previous studies have demonstrated an array of approaches for the use of lignin in nitrogen fertilizers. Ammoxidation binds ammonia chemically to lignin, but this causes limited nitrogen bioavailability to plants.¹⁷ Mulder et al.¹⁸ tested four different commercially available lignins as coatings for urea, but they found the lignin-based coatings ineffective since urea was completely released in water within one hour. García et al.¹⁹ produced coatings for urea granules from pine kraft lignin reinforced with resinous adhesives and linseed oil. While they were able to retard the dissolution of urea in water to some extent, this came at the price of either chemical modification or supplementation of lignin with costly materials.

The cost of the slow release fertilizer should not exceed the savings it provides by reduced fertilizer usage and increased harvest. Therefore, affordable lignin with well-suited properties to resist dissolution of associated urea would be needed. A key step that determines many properties of lignin is the isolation procedure. In addition to the well-known isolation by acidification of black liquor, lignin can be directly precipitated from alkaline solutions using polyvalent cations.²⁰ Amorphous kraft lignin is too brittle in lignin films,¹⁸ and thus additional carrier material is needed in the slow-release urea fertilizer formulation. Agricultural residues such as wheat straw represent low-cost and renewable materials and are thus attractive. Wheat straw was selected as a fertilizer carrier because compared to woody stems, straw contains a large empty internal volume and a hollow porous structure suitable for capturing urea and lignin. Straw has been tested as a

fertilizer carrier before, but mainly in approaches relying on chemical polymerization in the presence of milled or chemically processed straw.^{21–23} However, exploitation of straw or stalks of cereal plants with minimal processing has not been attempted in the production of slow release urea fertilizer systems.

The objective of the current study was to develop a slow-release urea formulation from renewable and low-cost materials. Wheat straw was selected as the fertilizer carrier and lignin as the binder component. Instead of using commercially isolated lignin, two fractions were isolated from pulping black liquor using acid and calcium precipitation, and compared for urea release. The differing aqueous solubility of these fractions (herein indicated as Acid-lignin and Ca-lignin), their surface areas, as well as chemical and morphological features suggested unique molecular-level structures. The effects of Acid-lignin and Ca-lignin on urea release in fertilizer systems is discussed.

EXPERIMENTAL SECTION

Materials

Softwood Kraft black liquor (KBL) from cellulose pulp production was obtained from Stora Enso Imatra mill (Finland). KBL at pH 13 contained 0.5% sodium hydroxide, 2% sodium sulfide and 68 g/L lignin (determined from the UV absorbance at 280 nm). Wheat straw was obtained as harvesting residues from Southern Finland in 2010. Calcium acetate monohydrate (99.4 %, J.T. Baker Chemicals, The Netherlands), sulfuric acid (95–98%, Sigma-Aldrich, Germany), and urea (>99%, Sigma, USA) were reagent grade chemicals.

Lignin isolation procedure KBL was used as the lignin source. Two procedures for precipitation of lignin were developed based on titration of KBL with either sulfuric acid or calcium acetate. In the first procedure, KBL (1000 mL) was precipitated by acidification at pH 3.7 by the addition of 4.2 M sulfuric acid (130 mL). In the second procedure, KBL (1000 mL) was precipitated by addition of 1 M calcium acetate (400 mL). The precipitated material in either of the procedures was recovered after centrifugation for 20 min at $10400 \times g$ using a Sorvall model RC 5B Plus centrifuge and a type GSA rotor. The supernatant was discarded and the pellet was air dried at 21 °C in a fume hood. The solid fractions were termed according to the precipitation method “Acid-lignin” and “Ca-lignin”, respectively.

Impregnation of straw with urea and lignin

A polytetrafluoroethylene cylinder mount with an internal diameter of 3.8 cm was filled with 7 g of 5 cm long sections from wheat straw. The lignin-urea melt was produced by mixing 50 g of lignin material in 50 g of urea at its melting point of 135 °C in a borosilicate glass decanter in a convection oven. The melt was stirred to homogeneity and cast over vertically aligned straw in the oven. The reference material for urea release tests was prepared by casting the straw with 25 mL of 800 g/L aqueous urea solution and allowing them dry overnight at 105 °C. The impregnated straw sections were cooled to room temperature, separated from each other and weighed to determine the material balances.

Urea and lignin leaching experiments

Leaching of urea or lignin from impregnated straw to deionized water at pH 7 was followed at time intervals. To determine the maximum urea release kinetics at 25 °C, two

straw sections (approximately 0.5 g) were subjected to 250 rpm agitation in a 250 mL conical flask containing 100 mL of deionized water. Tests at higher dry matter concentration (g dry matter/g total suspension) were conducted by subjecting a single 5 cm straw section and a given amount of deionized water to continuous rotation mixing in a screw cork glass tube inserted to a model SB2 rotator (Stuart), which enabled end-over-end mixing of vertically aligned tubes at 20 rpm speed. Urea was determined after 24 h mixing at 25 °C from the water phase. The urea release experiments were conducted with five replicates. Urea was analyzed with high-performance liquid chromatography (HPLC) using a system and analytical conditions as previously explained for monosaccharide analysis.²⁴

The release of urea from the lignin matrix was assumed to follow Fick's second law of diffusion with cylindrical geometry (Eq. S1). The diffusion coefficient D was determined by non-linear regression of the measured release of urea to the released concentration calculated from the time integral of the Fick's law, which was solved numerically using Matlab R2015a (Mathworks). The cylinder was discretized radially according to Özisic (1994)²⁵ (Eq. S2) and the time integral was solved in each section by using *ode15s*.

Characterization of lignin materials

Solubility of Acid-lignin and Ca-lignin (50 mg dry matter) in aqueous media was determined at 21 °C in screw cork glass tubes containing 5 mL of either deionized water, 0.05 M aqueous buffer solution at pH 4–10 (sodium acetate, sodium phosphate, $\text{NaCO}_3/\text{NaHCO}_3$, $\text{NH}_4\text{Cl}/\text{NH}_3$) or 0.1 M NaOH. The tubes were subjected to vertical rotation mixing at 20 rpm during 24 h. The final pH was measured and the lignin concentration was determined using the spectrophotometric method.²⁶ Absorbance at 280

nm was measured from the supernatant diluted in aqueous 0.1 M NaOH. Lignin concentration was calculated from Beer-Lambert law using Indulin AT lignin for calibration ($\epsilon=21.9$ L/g/cm). Lignin content of Acid-lignin and Ca-lignin was determined gravimetrically after the two-stage sulfuric acid hydrolysis procedure.²⁷ Solubility and lignin content were determined in duplicate.

Accessible lignin surface area was determined based on adsorption of the cationic dye Azure B on lignin.²⁸ In this method, lignin powder (50 mg on dry weight basis) was agitated at 25 °C in Azure B solution in 50 mL of 0.05 M Na-phosphate buffer (pH 7) for 24 h, and the remaining dye concentration was determined from the absorbance reading at 647 nm. The initial Azure B concentrations were 0, 0.1, 0.3, 0.5, 0.7, 0.8, and 1.0 g/L. A Langmuir isotherm was fitted to the mean values of duplicate adsorption data, and the maximum monolayer adsorption capacity was used to calculate the surface area covered by Azure B (1.30 m²/mg).

Calcium content of Acid-lignin and Ca-lignin was determined in three replicates by inductively coupled plasma-optical emission spectrometry (ICP-OES; Perkin Elmer ICP-OES, 7100 DV). Elemental analyses were performed in duplicate with Perkin-Elmer (PE) 2400 Series II CHNS/O Analyzer. For transmission electron microscopy (TEM), Acid-lignin and Ca-lignin were precipitated freshly from KBL and purified by four consecutive washes either with acidified water adjusted to pH 4 with 1 M HCl (Acid-lignin,) or three times with 0.01 M calcium acetate and once with deionized water (Ca-lignin). TEM was conducted using a FEI Tecnai 12 microscope operating at 120 kV. Imaging of 0.1% or 1% (w/w) lignin dispersions dried on carbon film support grid was done in a bright-field mode. Attenuated total reflection Fourier-transform infrared spectra of Calcium acetate as well as purified and crude Ca-lignin and Acid-lignin were recorded using a Mattson

3000 FTIR spectrometer (Unicam) with a GladiATR unit (PIKE Technologies) and EZ Omnic software (Thermo Nicolet Corp.). The total number of scans was 64 at the wavenumber range of 400–4000 cm^{-1} and at a resolution of 4 cm^{-1} . The spectra were subjected to automatic baseline correction. Dry matter content of various fractions was determined gravimetrically by drying samples at 105 °C to constant weight.

RESULTS AND DISCUSSION

Precipitation yields and composition of the raw materials

The objective of this work was to develop a new slow-release fertilizer system carrying urea in wheat straw impregnated with lignin. Approximately two-thirds of the same batch of wheat straw was previously found to consist of carbohydrates and less than one-fourth was lignin.²⁹ The isolation yields of the two unpurified lignins obtained from 1 L of KBL were 109.7 g (Acid-lignin) and 152.8 g (Ca-lignin) on oven dry basis. Acid-lignin showed a higher lignin content of 53% compared to 34% in Ca-lignin that contained 11% of calcium originating from the precipitation procedure, as it was nearly absent in Acid-lignin (Table 1). These values indicate precipitation of 85% (Acid-lignin) and 76% (Ca-lignin) of the original lignin in KBL, but it is obvious that both lignins contained salt residues from the precipitation procedures. In turn, there were notable differences in the elemental composition of Acid-lignin and Ca-lignin with 71% and 74% lower nitrogen and sulfur contents in the latter. While the excess sulfur in Acid-lignin originates from sodium sulfate formed in the acid-base titration of the black liquor, the higher nitrogen content suggests co-precipitation of denatured proteins or their degradation products in acidic conditions.

Table 1. Composition (g/100 g DM) of the materials used in this work.

| Material | C | H | N | S | O | Ca ^a | Lignin ^b | Carbohydrates |
|-----------------|------|------|------|------|------|-----------------|---------------------|-------------------|
| Wheat straw | 48.9 | 7.79 | 0.55 | 0.79 | 41.7 | 0.22 | 23.6 ^c | 66.3 ^c |
| Ca-lignin | 37.7 | 5.77 | 0.87 | 2.49 | 42.6 | 10.6 | 34.0 | na |
| Acid-lignin | 42.4 | 6.51 | 2.98 | 9.55 | 38.5 | 0.10 | 52.8 | na |
| Urea | 20.0 | 6.71 | 46.6 | - | 26.6 | - | - | - |
| Calcium acetate | 30.4 | 3.82 | - | - | 40.5 | 25.3 | - | - |

^a: by difference ^b: includes acid-insoluble lignin and acid-soluble lignin. ^c: data from literature for the same wheat straw material.²⁹

The differing purities of the two KBL-derived lignins can be explained by the higher amount of calcium acetate compared to that of sulfuric acid required to precipitate a given amount of lignin from KBL (Fig. S1). Both precipitation methods reached a similar level after which 10% of the original amount of lignin remained soluble, regardless of further supplementation of chemicals. This lignin fraction presumably consisted of low molecular weight fragments as it was acid-soluble and did not form insoluble chelation complexes with calcium. This finding is in agreement with the fact that chelation is a coagulation process in which only the high molecular mass fraction of kraft lignin is precipitated with calcium.²⁰ Using the elemental composition data, the number of Ca²⁺ ions bound to 100 structural C9 inter-units of lignin with assumed molecular weight of 182 g/mol is 142 compared to 33 reported earlier by Sundin.²⁰ In the current study, the crude lignin precipitate was not washed, leaving some soluble calcium acetate to the dried product. This decision was made to avoid costly separation processes, and because the presence of calcium may have a further advantage in soil by exchanging sodium and thus alleviating phytotoxic effects of elevated sodium concentrations on plants.³⁰ In 100 C9 units of kraft lignin, there are an estimated 15–20 carboxylic and 55–80 phenolic

groups.²⁰ Compared to this data, the value of 142 Ca²⁺ ions per 100 C9 units means that excess calcium was available for the chelation of the anionic groups of kraft lignin.

Mass uptake in impregnation of straw with urea and lignin

Impregnation of wheat straw sections with urea and lignin led to considerable mass uptakes, although the straw remained visually intact (Fig. S2). Straw absorbed 5.61 times its dry weight of 1:1 (w/w) mixture of urea and Ca-lignin. For Acid-lignin/urea, Acid-lignin/calcium acetate/urea, and urea alone the corresponding values were 3.30, 2.59, and 1.59. Therefore, Ca-lignin in molten urea was incorporated to the largest extent as is also evident when comparing the nitrogen contents of the fertilizer systems (Table 2). The urea content of Ca-lignin-containing straw was 46% compared to 61% in urea straw without lignin. The impregnation with urea alone was performed using concentrated aqueous solution instead of molten urea because of the HPLC detection of biuret, an unwanted reaction product of urea, in the latter case (data not shown). When lignin was dissolved in molten urea the formation of biuret was negligible for yet unknown reasons.

Table 2. Content of urea, lignin, nitrogen, and other material in fertilizer systems comprising urea only, wheat straw, urea-straw, and straw impregnated with lignin and urea.

| Fertilizer system | Content (g/100 g fertilizer system) | | | Nitrogen content (g/100 g fertilizer system) | |
|-------------------|-------------------------------------|---------------------|-------|--|-------|
| | Urea | Lignin ^a | Other | Urea | Other |
| Urea only | 100 | 0 | 0 | 46.6 | 0 |
| Wheat straw | 0 | 23.6 | 76.4 | 0 | 0.55 |
| Urea-straw | 61.4 | 9.1 | 29.5 | 28.6 | 0.21 |

| | | | | | |
|--|------|------|------|------|------|
| Ca-lignin-urea-straw | 45.5 | 38.5 | 15.9 | 21.2 | 0.41 |
| Acid-lignin-urea-straw | 41.6 | 40.7 | 17.8 | 19.4 | 1.2 |
| Acid-lignin/calcium acetate-urea-straw | 36.1 | 26.4 | 37.5 | 16.8 | 0.7 |

^a.Total lignin content (extraneous lignin added to the amount of lignin from straw)

Release of urea and lignin from impregnated straw in water

Urea dissolves rapidly in water within a few minutes when stirred in dilute water suspension. When urea was impregnated into the porous straw matrix 87% of urea dissolved after shaking in excess water for 1 h (Fig. 1). Impregnation of straw with lignin in molten urea caused a marked reduction in the release rate of urea. During 1 h 70% of urea was dissolved from Ca-lignin and 79% from Acid-lignin. To test whether this difference was caused mainly by calcium ions, Acid-lignin was supplemented with calcium acetate at an equal 10.6 % (w/w) calcium content as found in Ca-lignin (Table 1). It turned out that the dissolution of urea from straw sections impregnated with calcium added *ex-situ* was similar compared to that of straws impregnated with Ca-lignin (Fig. 1). It seemed, therefore, that in addition to complex formation with lignin, Ca²⁺ ions interact also between anionic groups of hemicelluloses and lignin.

Although the process of urea release includes dissolution and water penetration into the material, the kinetics of the release can be simplified as a diffusive process, following the Fick's second law of diffusion. The adapted diffusion model overestimated the release of urea to a certain degree, but showed coefficient of determination values $0.90 < R^2 < 0.96$ (Fig. 1 and Table S1). The diffusivity coefficient D provides a measure for the retention of a solute inside a solid matrix and can be applied for the prediction of urea

release for different particle sizes and geometries. Significantly lower diffusivities ($0.0017\text{--}0.0018\text{ m}^2/\text{s}$) and thus higher urea retentions were observed for both calcium containing lignins compared to Acid-lignin ($0.0027\text{ m}^2/\text{s}$), while the urea-straw showed the highest diffusivity of $0.0070\text{ m}^2/\text{s}$, in accordance with the most rapid release of urea.

It appears that the release of urea from the fertilizer systems was obstructed by additional factors since the dissolution of urea was slower than predicted from the simplified diffusion model. To gain more insight to this phenomenon, the dissolution of urea from the straws impregnated with Ca-lignin and urea was followed for 72 h. In this case, the dissolution of urea increased slowly from 4 h and reached 104% (standard deviation 4.6%) from straw impregnated with Ca-lignin after 72 h immersion in water (Fig. S3). From other fertilizer systems, the 24 h urea release percentages were 95.2% (urea-straw), 93.2% (Acid lignin-urea-straw), 96.0% (Acid lignin-calcium acetate-urea-straw), and the pooled standard deviation was 5.4%. These high urea release values at neutral pH suggest that the binding mechanism of urea was solely physical. Such a slow release rate at high moisture conditions suggests dissolution limited by steric effects such as diffusion from the micro- and nano-scale pores present in straw.³¹

In consequence, the impregnation method offered beneficial effects compared to coating of urea with lignin reported earlier. Coating urea with lignin resulted in complete release of urea within one hour, despite the chemical modification of the lignin-based coatings.¹⁸ Based on the visible color changes during the prolonged incubations it was obvious that partial dissolution of lignin occurred in addition to the dissolution of urea. Increasing UV absorbance confirmed that the initial release of lignin during the first hour was followed by a slower phase (Fig. 2). However, the dissolution of lignin was much slower compared to that of urea release. After 7 days of incubation, the amount of Acid-lignin released was

two times as much as that of Ca-lignin and three times compared to the dissolution of Acid-lignin supplemented with calcium acetate. Therefore, the slow release rate of lignin holds potential to further improve the fertilizer system. For instance, it could be advantageously used for the slow release of a second fertilizer component chemically bound to lignin.

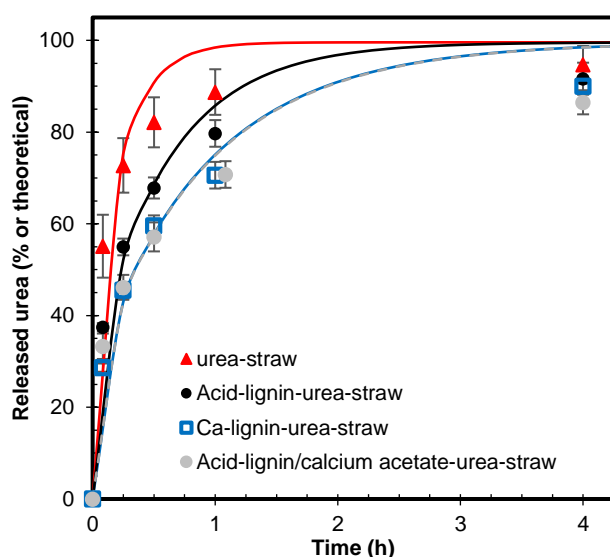


Figure 1. Urea dissolution kinetics from straw impregnated with urea only or lignin in molten urea (1:1 w/w). Two straw sections (5 cm) were agitated at 250 rpm in 0.5% suspension in deionized water at 25 °C. Curves represent Fickian diffusion models.. The error bars indicate one standard deviation from the mean of five replicates.

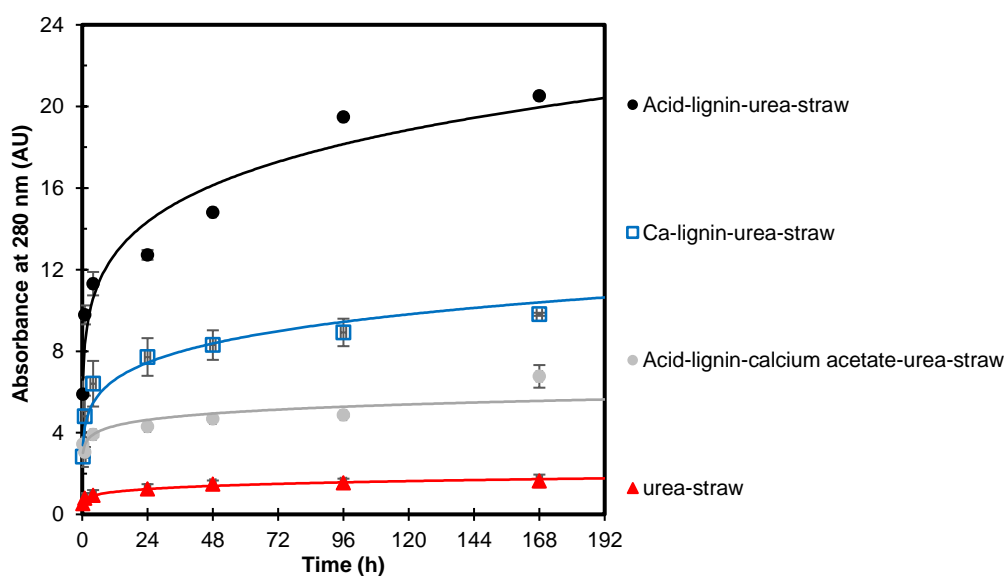


Figure 2. Lignin dissolution kinetics from straw impregnated with urea only or lignin dissolved in molten urea (2.5 cm straw sections). Absorbance in the liquid phase at 280

nm is expressed relative to impregnated straw dry weight. Curve fitting parameters are given in the SI (Table S2). The error bars indicate one standard deviation from the mean of two replicates.

The previously discussed urea and lignin release tests were carried out in excess water. The moisture content of soils is typically much lower and can be expected to vary considerably in response to the environmental conditions. The extent of dissolution of urea in 24 h was determined at varying straw dry matter contents. It is evident that the dissolution of urea was inversely proportional to the dry matter content regardless whether additional lignin was present or not (Fig. 3). The extent of water absorption into Ca-lignin was so low that liquid samples could be collected from the suspension at 72% dry matter concentration. The corresponding proportion of urea dissolved was 42% which is comparable to the value of 45% obtained in 24 h from synthetically produced urea fertilizer system placed in soil at 70% dry matter content.²² Compared to these fertilizer systems requiring a multi-step chemical synthesis,²² the simple impregnation of urea in straw with lignin in the present work marks a significant advancement in the development of affordable slow release formulations.

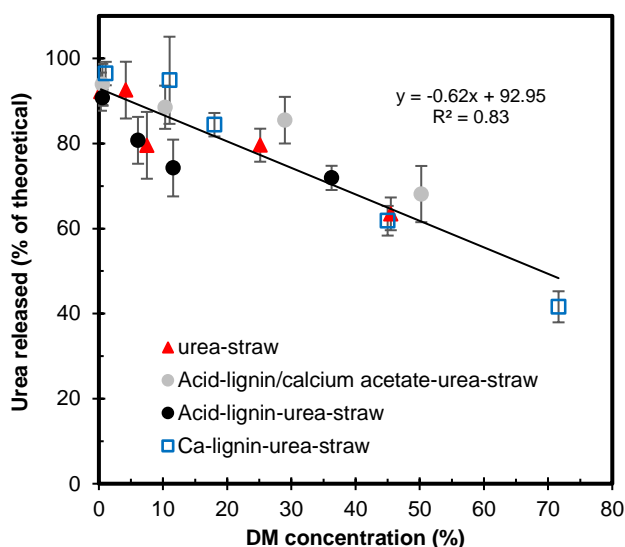


Figure 3. Effect of dry matter concentration on the dissolution of urea from straw impregnated with urea only or lignin in molten urea (1:1 w/w) during 24 h at 25 °C. The error bars indicate one standard deviation from the mean value of five replicates.

Understanding the effects of lignin on retarding dissolution of urea

Lignin is known to provide water resistance to lignocellulosic materials such as cellulosic fibres.³² Properties of the two lignins obtained from the pulping spent liquor were studied to better understand how they affect water penetration and dissolution of urea from the impregnated straws.

Ca-lignin has pH-independent solubility in aqueous media

The aqueous solubility of the two lignins was studied over a broad pH range to gain understanding of their stability in the fertilizer systems. The solubility tests were conducted at room temperature during 24 h in continuous rotation mixing, and the final pH of the suspension was recorded. Aqueous solubilities of 10–30% were similar between the two lignins at pH 4–7 (Fig. 4). The solubility of Acid-lignin increased as a function of pH (starting pH 4) and plateaued at 90% after pH 9.5. The solubility of Ca-

lignin deviated from this typical pH-dependent solubility of kraft lignin. A particularly interesting difference was observed at alkaline pH conditions, where the extent of dissolution of Ca-lignin depended on the type of counterion present in the aqueous buffer solution. In sodium carbonate/sodium bicarbonate buffer at pH 10, the solubility of Ca-lignin was 92% which was comparable to the value of Acid-lignin. In contrast, in ammonium chloride/ammonia buffer at pH 10 the solubility of Ca-lignin was only 28%. This is explained by the formation of insoluble calcium carbonate precipitate from Ca-lignin in the carbonate buffer, which is in contrast to the case of $\text{NH}_4\text{Cl}/\text{NH}_3$, which did not show any precipitation. Therefore, the underlying reason for the lower dissolution of Ca-lignin in the ammonium chloride/ammonia buffer is the presence of soluble calcium chloride that maintained calcium chelation of lignin. Because of their generally higher pKa values compared to aliphatic carboxyl groups,³³ the phenolic hydroxyl groups had the largest effect on lignin solubility above pH 6. This result further confirms that anionic phenolate ions promote solubility of lignin in alkaline media, as previously shown using methylation and thioacidolysis.³⁴ Therefore, the water-solubility of Ca-lignin can be controlled by the calcium ion concentration and the type of anionic counter ions present in water.

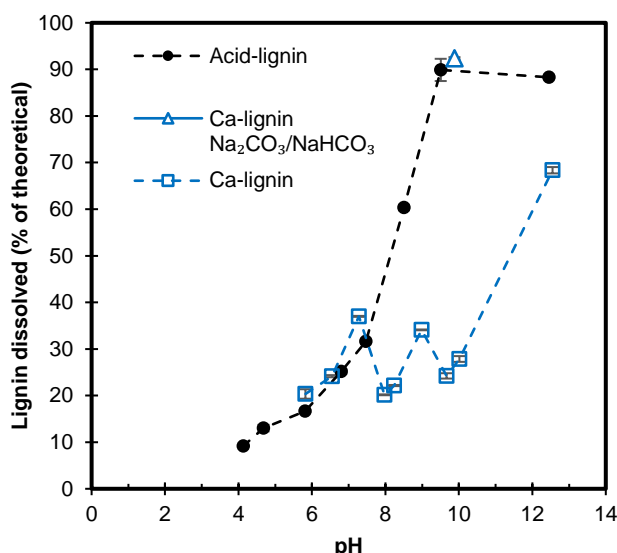


Figure 4. Aqueous solubility of Acid-lignin and Ca-lignin. Lignin materials in deionized water, pH buffer solutions, or 0.1 M NaOH were subjected to continuous end-over-end agitation during 24 h at 21 °C. The error bars indicate average deviation from the mean value of duplicate experiments.

Comparison of morphological and chemical structures of Ca-lignin and Acid-lignin

To gain insight to the contrastive solubilities and performances in the urea fertilizer systems, Ca-lignin and Acid-lignin were further characterized. For this, kraft black liquor was precipitated in small scale either with 1 M calcium acetate or 4.2 M sulfuric acid and the obtained materials were washed free of soluble materials. The purified Ca-lignin and Acid-lignin were subjected to TEM analysis to visualize the materials from micrometer to nanometer scale. Both lignins formed three-dimensional precipitates that had agglomerated together as larger interpenetrating clusters (Fig. S4 a, d). Distinct features of approximately 10–20 nm in Acid-lignin (Fig. S4 g, h) resembled those of lignin precipitated from anhydrous tetrahydrofuran solution by iron (III) isopropoxide.³⁵ At higher magnification, Ca-lignin appeared more "flaky" compared to denser Acid-lignin at roughly similar magnifications (Fig. S4 b, e). Though these differences may result partly from the heterogeneity of the lignin suspensions, the fine structure of Ca-lignin was clearly less compact than Acid-lignin (Fig. S4 c, f). It is obvious that the morphology of

the lignin materials changed upon dissolution in molten urea, but the TEM investigation nevertheless showed interesting fine structures of lignins obtained through different precipitation routes.

Surface area is an important property affecting the release of urea. The accessible surface area of lignin was determined by adsorption of cationic dye Azure B on unpurified Acid-lignin and Ca-lignin to compare their functionality in slowing down the dissolution of urea from impregnated straw. The dye adsorption on kraft lignin occurs into sterically unhindered anionic and acidic functionalities, mainly phenolic hydroxyls and carboxylic groups.²⁸ When related to the lignin content of the materials, Ca-lignin showed a higher maximum adsorption capacity compared to Acid-lignin (Fig. S5). In terms of specific surface area (SSA), Ca-lignin showed SSA of 997 m²/g-lignin compared to 881 m²/g-lignin of Acid-lignin. This means that Ca-lignin was more accessible to adsorption of Azure B than Acid-lignin, in turn, in agreement with the observations from the TEM analysis, this suggests a less dense structure that did not hinder the anionic sites as much as in a more aggregated Acid-lignin. To our knowledge, no published surface area data exist for isolated lignins based on the dye adsorption methodology. Meanwhile, it is known that the abundant phenolic hydroxyl groups are the major adsorption sites for Azure B in lignin.²⁸ The higher accessible surface area of Ca-lignin compared to that of Acid lignin is speculated to be a result of calcium chelation of phenolate ions, which prevented the formation of dense molecule clusters and facilitated adsorption of Azure B through cation exchange.

Stabilization of Ca-lignin, its partially pH-independent solubility, and distinct morphological structure compared to Acid-lignin prompted to record infrared spectra of the purified lignins. The purification procedure, which was the same one conducted

before the TEM investigation, was successful as judged from the absence of prominent bands of calcium acetate at 460 cm^{-1} and $600\text{--}670\text{ cm}^{-1}$ in the purified Ca-lignin (Fig. S6). The spectra of purified lignins were compared to those of industrial kraft lignins, aiming to understand how the chelation affected the functional properties of Ca-lignin. The assignment of the lignin bands was according to the literature.³⁶ Overall, the spectra of purified Acid-lignin and Ca-lignin differed notably from each other whereas the spectra of Acid-lignin and the reference lignins (Indulin AT and Lignoboost) were nearly identical (Fig. 5). Purified Ca-lignin showed a lower intensity for the bands at 1592 cm^{-1} and 1510 cm^{-1} compared to that of purified Acid-lignin. These bands arise from aromatic skeletal vibrations and could be stabilized in Ca-lignin by $\pi\text{--}\pi$ interactions of aromatic rings that were organized in chains as a result of calcium chelation of phenolic hydroxyls. The band at 1365 cm^{-1} has been assigned to aliphatic C–H stretch in methyl group (not MeO) and phenolic OH, the latter being more abundant in lignins. This band was more distinct in Acid-lignin and the reference lignins compared to Ca-lignin in which the phenolate ions were chelated with calcium. In addition, the band at 1268 cm^{-1} is typical for guaiacyl ring stretch and was weaker in Ca-lignin than in Acid-lignin. Further suggestion for the presence of planar chelation chains in Ca-lignin came from the lower intensity of the aromatic C–H in-plane deformations (1030 cm^{-1}) and stronger C–H out-of-plane deformations of guaiacyl units at 870 cm^{-1} .³⁷ In-plane C–H deformations in guaiacyl units would be hindered by the calcium atom in the proximity of the C3 position of the aromatic ring whereas out-of-plane deformations would show the opposite trend (Fig. 6). Taken together, these results suggest that chelation of Ca-lignin led to a preferential planar chain orientation of lignin molecules dictated by the coordination with the bivalent calcium cations (Fig. 6).

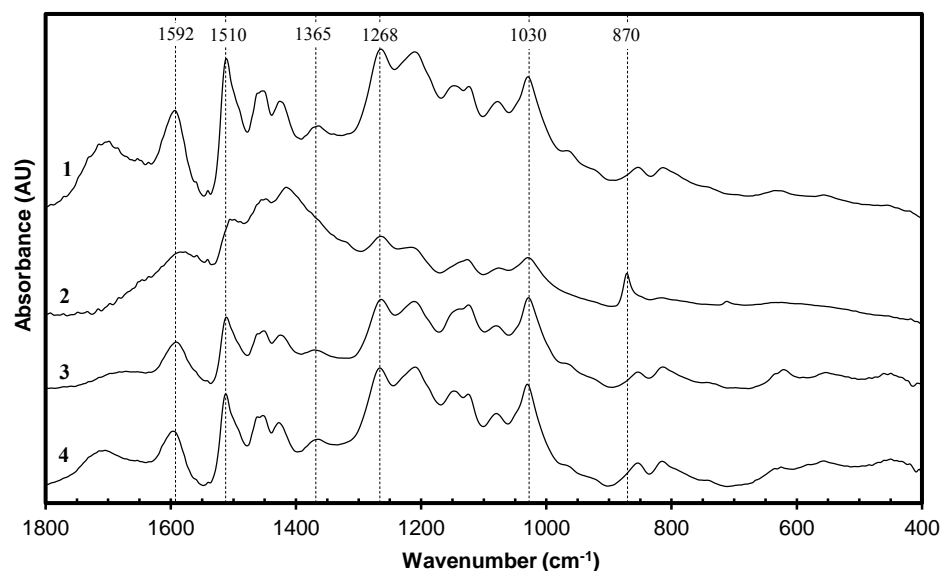


Figure 5. The fingerprint region of infrared spectra of 1) purified Acid-lignin 2) purified Ca-lignin 3) Indulin AT lignin 4) Lignoboost lignin.

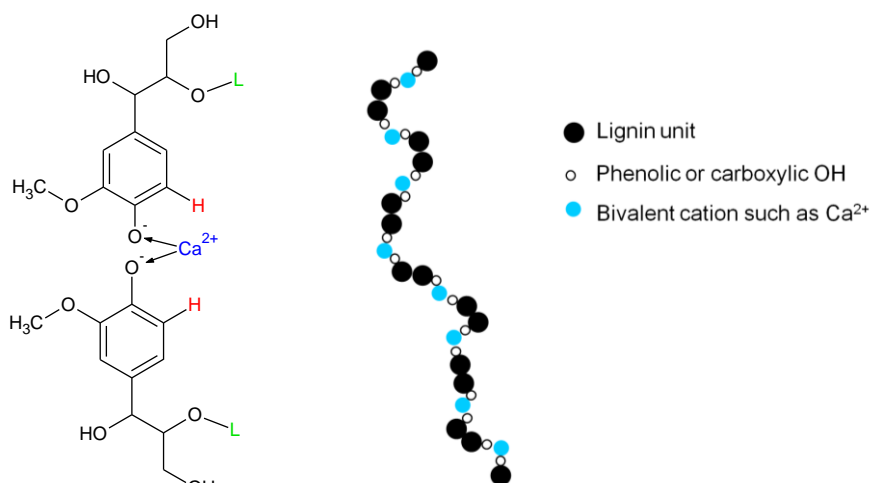


Figure 6. The proposed in-plane orientation of the aromatic rings of guaiacyl units as a result of chelation with a calcium ion. Only the aromatic hydrogen ions adjacent to the calcium ion are shown. L=lignin. The formation of planar chains results and leads to precipitation of lignine from alkaline solution.

Applicability of acid and calcium acetate in lignin isolation

To produce lignin fractions with contrastive properties for the fertilizer systems, precipitation of kraft black liquor was undertaken separately with sulfuric acid and calcium acetate. The benefit of using calcium acetate is that lignin thus obtained is insoluble at alkaline pH since only a slight drop in pH resulted from this precipitation process. As demonstrated in the present work with the fertilizer systems, lignin with low

or controlled water-solubility would be useful in applications in which water-resistance is needed. Industrially, carbon dioxide supplementation to kraft black liquor is used to reduce the pH to about 10 and precipitate lignin for subsequent washing with water containing sulfuric acid.³⁸⁻⁴⁰ In the Lignoboost[®] process the operation costs is mainly related to the consumption of chemicals.⁴¹ Alkaline oxidation is known to convert carbohydrates partially into organic acids and reduce pH and molar mass of lignin.⁴² Consequently, less acidification chemicals are needed. LignoForce System subjects concentrated black liquor to such initial oxidative treatment that avoids formation of reduced sulfur compounds in the successive lignin precipitation with carbon dioxide.⁴³ Calcium acetate is more expensive compared to carbon dioxide or sulfuric acid, but opportunities might exist in a combinatorial process. After the initial pH reduction by carbon dioxide, less calcium acetate would be needed for the formation of insoluble calcium chelation complexes.

CONCLUSIONS

Kraft pulping spent liquor was used as a low-cost source of lignin for the preparation of slow release urea fertilizer systems in wheat straw matrix. Calcium precipitation formed Ca-lignin with contrastive functional properties compared to Acid-lignin obtained by acidification. The slow dissolution of urea from straw impregnated with Ca-lignin demonstrated a simple and affordable fertilizer system. Chelation of phenolic hydroxyl groups of lignin with calcium was responsible for the reduced water-solubility, higher surface area, and less densely packed nanoscale morphology of Ca-lignin compared to corresponding properties of Acid-lignin. Further investigations could be undertaken to exploit the polyelectrolyte property of lignins for instance by cationic chelation for the fabrication of structured materials.

ASSOCIATED CONTENT

Titration curves of KBL with sulfuric acid or calcium acetate, Photographs of wheat straw sections after impregnation with urea and lignin, Finite difference solution to the diffusion equation, Dissolution kinetics of urea from straw impregnated with Ca-lignin, Transmission electron micrographs of Ca-lignin and Acid-lignin, Azure B adsorption isotherms of Acid-lignin and Ca-lignin, Infrared spectra of calcium acetate, unpurified Ca-lignin, and purified Ca-lignin, parameters of the polynomic models of lignin dissolution from impregnated straw (PDF).

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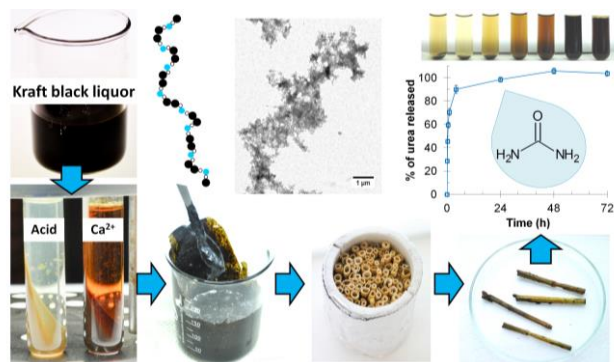
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TOC graphic:

Manuscript title: Calcium chelation of lignin from pulping spent liquor for water-resistant slow-release urea fertilizer systems



Synopsis: Calcium chelation of alkaline pulping spent liquor provides Ca-lignin with low water-solubility and beneficial effects in slow-release urea fertilizer systems