

Original article

Isolation and characterisation of cellulose nanocrystals from kudzu (*Pueraria montana* var. *lobata*): a sustainable packaging materialSneh Punia Bangar,¹  Reid Love,¹ William Scott Whiteside,^{1*}  Hansol Doh,² Duncan O. Darby¹ & William S. Bridges¹¹ Department of Food, Nutrition, and Packaging Sciences, Clemson University, Clemson 29634, USA² Department of Food Science and Biotechnology, Ewha Womans University, Seodaemun-gu, Seoul 03760, Republic of Korea

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Summary Flexible packaging has become indispensable in the food industry due to its numerous advantages. However, it has certain drawbacks, such as limited recyclability and environmental contamination. While efforts are underway to develop more sustainable and eco-friendly packaging options, the food industry's reliance on flexible packaging made from unsustainable petro-based plastics is expected to continue in the foreseeable future. Therefore, more sustainable alternatives that do not sacrifice performance are required. One such option is to use nanocellulose, especially cellulose nanocrystals (CNCs), which can be reinforced into biopolymer to enhance mechanical and barrier properties. The present study aims to isolate kudzu CNCs by depolymerisation, bleaching, acid hydrolysis with 51% sulfuric acid, and mechanical dispersion. Thermogravimetric analysis (TGA) was conducted to assess the thermal stability of kudzu fibres. Further, the FTIR spectra were measured at wave number ranged 4000–500 cm⁻¹, which showed changes in the structure after treatments and confirmed the removal of lignin and hemicellulose from the cellulose. The isolated CNCs were found to have lengths and diameters of 285.01 ± 150.33 nm and 42.78 ± 13.26 nm, respectively, with an aspect ratio of 6.43 ± 1.78. This study showed that kudzu CNCs are a suitable reinforcing material for nanocomposites.

Keywords Acid hydrolysis, cellulose nanocrystals, fibres, kudzu, packaging.

Introduction

Flexible packaging provides several benefits for the food industry, including protection, preservation, and convenience, and it is unlikely that the industry reliance on this packaging will diminish in the foreseeable future. Worldwide plastic production is currently at 390.7 million metric tons and is projected to increase by 10.8 million tons per year over the next decade (Brandon & Criddle, 2019). This growth rate far exceeds the estimated annual recycling capacity of 0.8 million tons, leading to growing public concern and dwindling landfill space. Several biopolymer-based packaging has been developed as an alternative to petroleum-based packaging to address this issue. However, performance, processing, and costs are some issues that could arise if biopolymers are to replace synthetic polymers on a large scale (Rodriguez-Perez

et al., 2018). Moreover, biopolymers have difficulty achieving the desired mechanical and thermal stability for processing and the water and oxygen barrier needed for consumer product protection (Punia Bangar *et al.*, 2022a). As a solution, reinforcement of biopolymers with fibers can improve the functionality of biopolymers (Bangar *et al.*, 2023).

A sustainable and cost-efficient approach is to add cellulose/nanocellulose to the biopolymers (Cruz *et al.*, 2022; Punia Bangar *et al.*, 2022b). In its native form, cellulose is often found in a lignin and hemicellulose matrix. To extract cellulose, alkaline agents and weak acids are used to solubilise the non-cellulosic fractions (Doh *et al.*, 2020). The conversion of cellulose to its nanoform (CNCs) has several packaging advantages due to its high elasticity and tensile strength. Moreover, due to their unique properties, cellulose nanocrystals (CNCs) have the ability to function as nucleating agents and enhance the crystallinity of polymers (Rahighi *et al.*, 2021). This characteristic

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makes CNCs an appealing choice for reinforcing biopolymers. Therefore, amorphous and noncellulosic fractions are isolated from cellulose to leverage cellulose functionality fully.

This study aimed to extract CNCs using kudzu (*Peararia montana* var. *lobata*). Kudzu is an invasive plant species native to Asia but has become a major problem in the southeastern United States as “the vine that ate the South” (Gulizia & Downs, 2019). It was originally introduced to the US in the late 1900s to control erosion and provide forage for livestock. Kudzu is known for its rapid growth and ability to overtake and strangle the vegetation in its surrounding area. Considering its exceptionally rapid propagation, kudzu is now widely considered an agricultural nuisance. Local populations always welcome its removal and harvesting in the United States for alternative purposes. As per the literature review, isolating CNCs from kudzu has never been documented. Thus, this research aims to isolate and characterise the aerial and vine regions of kudzu for its packaging applications.

Materials and methods

Kudzu collection

Kudzu leaves and vines were harvested from a heavily infested area in Clemson, South Carolina, USA. The leaves were washed, dried, and grind into a fine powder using a commercial blender (Magic Bullet, USA). The kudzu vines went through a retting process. The vines were cut into 1 × 15 cm lengths, followed by water soaking for 4 days. Later, the outer layers of fibres were peeled off and oven dried for 72 h at 30 °C. Later, the fibres were grind into a fine powder using the blender.

Isolating cellulose nanocrystals

The cellulose nanocrystals (CNCs) extraction process from kudzu was completed in 4 steps, including depolymerisation, bleaching, acid hydrolysis, and mechanical dispersion. The pulverised fibres were mixed with 0.2 M HCl at a 1:15 ratio (w/v) with a magnetic stirrer for 2–3 h at 30 °C to begin the depolymerisation step. The fibre suspensions were water-washed using a centrifuge (Avanti® 144 JXN-30, IN, USA) at 15 000 g for 5–10 min to discard the supernatant. At 7 pH, the leaves and vine residues were mixed with distilled water separately at a 1:60 ratio (w/v), then adding 4% NaOH until 10 pH was reached. The solutions were placed on a hot-plate magnetic stirrer for 3 h at 70 °C. Later, suspensions were water-washed using the centrifuge at 15 000 g for 10–15 min to remove excess NaOH. Finally, the residues were oven dried for 3 days at 65 °C.

For the bleaching process, the residues were subjected to 10% KOH at a 1:20 (w/v) ratio for 3 h at 30 °C. At this stage, leaves were damaged, but vine residue processing continued. The vine residues were mixed with 6.5% NaClO (w/v) for 12 h at 15 °C, followed by water washing using the centrifuge for 10 min. Further, the pH was adjusted to 5 using glacial acetic acid and allowed to stir for 2 h to remove hemicellulose. The 2nd bleaching was done with 30% H₂O₂ for 70 min at 80 °C to remove lignin and remaining hemicellulose. The solution was water washed using the centrifuge to remove the supernatant. The bleached fibres were used for acid hydrolysis.

The bleached fibres were converted to nanosize during the acid hydrolysis by removing amorphous areas. The bleached fibres were mixed with 51% H₂SO₄ with continuous stirring for 30 min at 45 °C with a 1:15 (w/v) ratio. Further, the solution was water-washed multiple times and centrifuged to get CNCs suspension. The pH was adjusted to 7 using 4% NaOH and subjected to mechanical dispersion *via* an ultrasonicator (Q700 Sonicator, Connecticut, USA) at 30% amplitude for 15–20 min. The suspension was immediately lyophilised by a commercial freeze dryer to avoid aggregation of CNCs and yield a fine powder.

Characterisation of kudzu CNCs

Zeta potential of cellulose nanocrystals

Zeta potential of kudzu CNCs was measured by a Malvern Zetasizer Nano-Z (Malvern Instruments, Worcestershire, UK). Measurement parameters were set as follows: refractive index, 1.470; absorption, 0.100; dispersant (distilled water) refractive index, 1.330.

Morphological analysis

The morphology of CNCs was conducted using transmission electron microscopy (H-9500, Hitachi, USA). The dimensions of the CNCs were calculated with ImageJ software. Ten different CNCs were examined for length (*L*) and diameter (*D*) to measure the average aspect ratio.

FTIR analysis

A milligram of untreated, bleached, and acid-hydrolysed vines was individually kept on the crystal beam of the attenuated total reflectance (ATR)-FTIR spectrometer (Nicolet iS10, Thermo Fisher Scientific, USA). Parameters used in the measurements were: resolution 4 cm⁻¹, spectral range 500–4000 cm⁻¹, and scans 128.

XRD analysis

The untreated, bleached, and acid-hydrolyzed vines were examined for XRD analysis to determine crystallinity index (CI). Using a scanning ratio of 2 degrees min⁻¹ and a range of 5–40 degrees, the CNCs were radiated

with copper K- α (wavelength = 1.5418 Å) using a 40 kV voltage and 40 mA current. The crystallinity of the CNCs was determined using the Segal method (1959).

$$CI (\%) = \frac{I_{200} - I_{am}}{I_{200}} \times 100$$

where I_{200} = peak intensity of the plane ($2\theta \cong 23^\circ$), and I_a = peak intensity of the amorphous fraction ($2\theta \cong 19^\circ$).

Thermogravimetric analysis

The untreated, bleached, and acid-hydrolysed samples were examined for thermogravimetric analysis (TGA; Hi-Res TGA, TA, USA). 2–10 mg samples were placed on an aluminium pan and heated in an atmosphere of nitrogen. The samples were heated over 30–600 °C at a heating rate of 10 °C min⁻¹. Derivative thermogravimetric (DTG) data was generated with the formula:

$$\text{Derivative Weight} \left(\frac{\% \text{weight change}}{\text{min}} \right) = - \frac{\frac{m_i - m_{i-1}}{m_o} \times 100}{t_i - t_{i-1}}$$

where m_i = mass of the sample at the i th index, t_i = time (in min) at the i th index, and m_o = mass of the original sample before heating.

Statistical analysis

Data analysis with a t -test was used to determine any statistically significant difference between the aspect ratios, lengths, and diameters of the respective CNCs ($P \leq 0.05$).

Results and discussion

After the depolymerisation step, the CNCs yield of the kudzu leaf was too low to continue with further treatment. Therefore, only CNCs produced from the vine were analysed.

Morphology analysis

The isolated CNCs were found to have lengths and diameters of 285.01 ± 150.33 nm and 42.78 ± 13.26 nm, respectively, with an aspect ratio of 6.43 ± 1.78 . The zeta potential of kudzu CNCs was -39.6 mV, which indicated that nanoparticles are stable in the suspension. The TEM image of isolated CNCs is shown in Fig. 1. The average aspect ratio of the kudzu vine CNCs was comparable to the sargassum seaweed but lower than that of the kombu seaweed when compared to the Doh *et al.* (2020) samples (who employed a similar approach; Table S1). Further, the greater length of the kudzu vine CNCs compared with seaweed CNCs could be attributed to a high degree

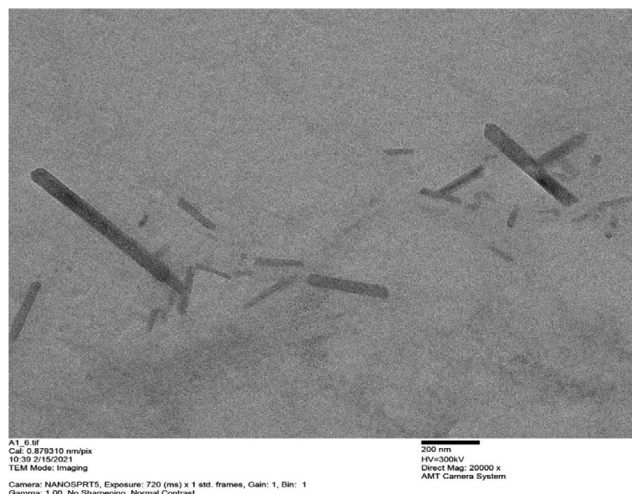


Figure 1 TEM image of kudzu vine cellulose nanocrystals (CNCs).

of polymerisation (DP) of kudzu fibres (Vanderfleet & Cranston, 2021). In contrast, the lower aspect ratio of kudzu CNCs (6.43) to the kombu seaweed CNCs (11.13) could be due to the larger diameter of the kudzu vine CNCs. The decreased hemicellulose content of the kudzu vine compared to seaweed sources may explain the bigger diameter. This finding is consistent with earlier research. Mozdyniewicz *et al.* (2016) discovered that decreasing hemicellulose content was linked to increased fibril width.

FTIR analysis

The FTIR technique was carried out to study the functional groups in untreated, bleached, and acid-hydrolysed kudzu vine and to examine the changes that occurred due to various treatments. The untreated kudzu vines displayed 3 peaks specific to lignin and hemicellulose, which were diminished in the bleached vine spectrum and were absent in the acid-hydrolysed vine spectrum (Fig. 2). The first peak at 1236 cm⁻¹ in the untreated vine represents aryl-alkyl ether linkages in lignin, which vanished in samples that had been bleached and acid hydrolysed, proving that lignin had been removed. Second, the untreated vine showed a band between 1500–1600 cm⁻¹, suggesting the presence of lignin aromatic rings. The bleached and acid-hydrolysed samples did not show this band. The band at 1507 cm⁻¹ ascribes aromatic ring vibrations, and the absence of any band in this range confirms lignin removal (Luzi *et al.*, 2014). Lastly, the untreated vine showed a clear peak at 1733 cm⁻¹, whereas the vine that had been bleached showed significantly less, and the vine that had been acid hydrolysed showed no peak. The acetic and uronic acid ester groups that are specific to hemicellulose or the carboxylic acid ester

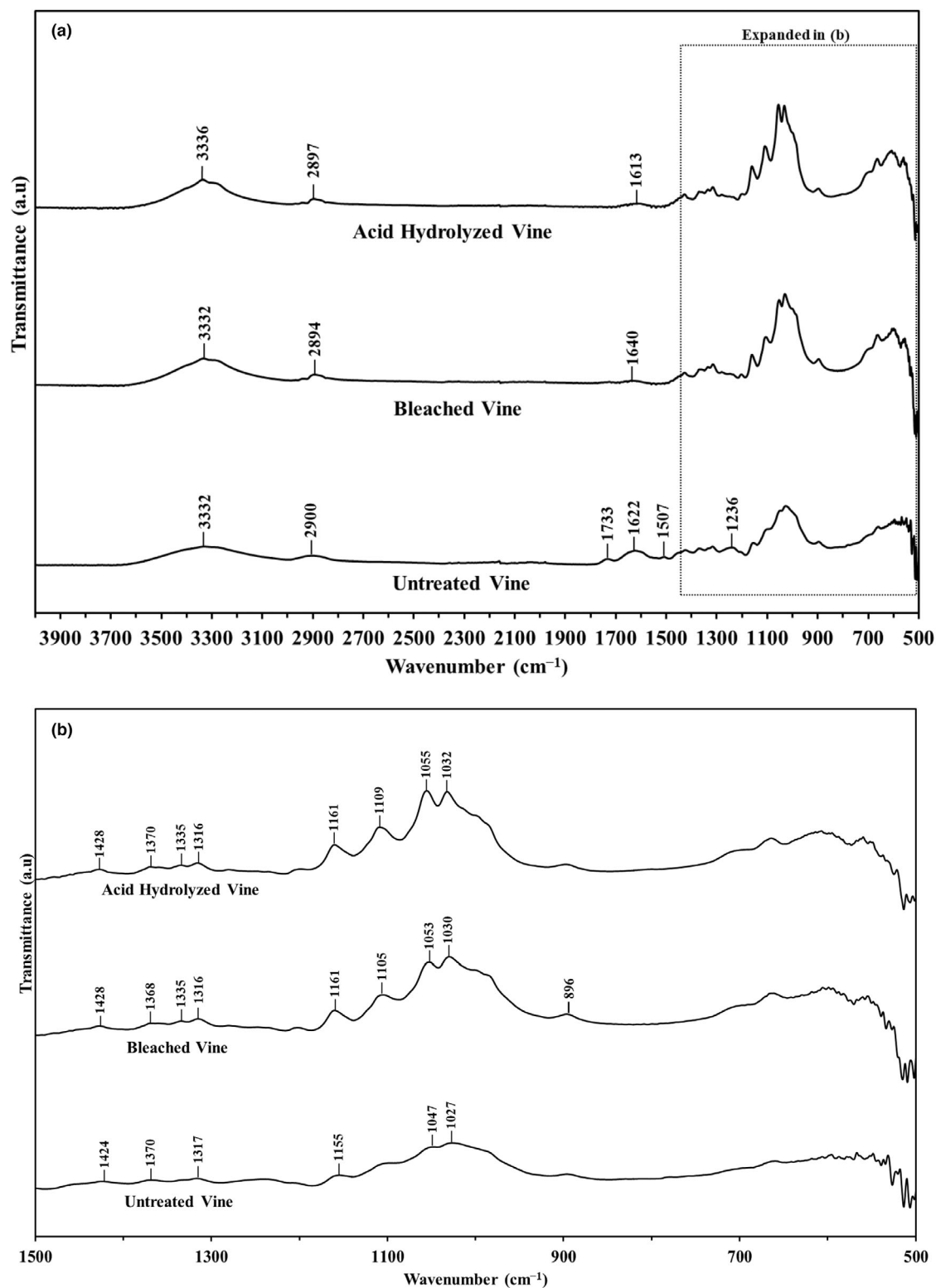


Figure 2 FTIR spectra of (a) the untreated vine, bleached vine, and acid hydrolysed vine in the 4000–500 cm^{-1} region, and (b) expanded region between 1500 and 500 cm^{-1} .

groups in the ferulic and p-coumaric acids that are characteristic of lignin and hemicellulose could be responsible for these peaks (Chirayil *et al.*, 2014; Chandra *et al.*, 2016). The disappearance of these peaks demonstrated the extraction of cellulose from the hemicellulose and lignin in the cell wall matrix.

The FTIR results shed light on the cellulose structure as well. The peak observed at 896 cm^{-1} was attributed to the deformation of β -glycosidic linkages linking the glucose rings of cellulose (Chirayil *et al.*, 2014; Luzi *et al.*, 2014). Broadband with relatively strong intensity indicated the presence of cellulose I structure, while weak intensity was associated with relatively amorphous cellulose (Fig. 2b). Based on this peak intensity, the acid hydrolysed and bleached vine samples showed a more crystalline structure than untreated vine. The change from an untreated state of 1424 cm^{-1} to a treated state of 1428 cm^{-1} was indicative of a C-H2 scissoring motion. The treated state showed a strong peak at 1428 cm^{-1} , indicating cellulose I, and a less intense peak at around 1420 cm^{-1} , indicating a decrease in crystallinity. For the treated samples, the band at 1161 cm^{-1} was associated with the C-O-C stretching of the antisymmetric glycosidic linkages in cellulose I, whereas in the untreated vine, this band was shifted to 1155 cm^{-1} , indicating a more amorphous structure. The increased intensity of the 1161 cm^{-1} peaks of the treated samples compared to the weak 1155 cm^{-1} peak of the untreated sample explained lignin and hemicellulose removal, leading to the treated samples falling into more compact and crystalline structures than the more heterogeneously structured untreated samples (Chandra *et al.*, 2016). Furthermore, the peaks in the $1047\text{--}1055\text{ cm}^{-1}$ range further demonstrated the cellulose isolation in the treated samples, with its increasing intensity indicating increasing cellulose and decreasing non-cellulosic components (Doh *et al.*, 2020). The peak at 1032 cm^{-1} showed the vibrations of the C-O stretching in cellulose, which was most pronounced in the acid-hydrolysed vine (Zhuang *et al.*, 2020). However, it is important to be cautious while interpreting conclusive results as the spectra of cellulose, lignin, and hemicellulose can overlap in certain regions, such as around 1032 cm^{-1} , which could indicate the presence of residual lignin in the bleached and acid hydrolysed vine. Hence, FTIR analysis cannot completely rule out the presence of residual lignin.

The passage implies that having a high degree of hydrophobicity for packaging materials is beneficial (Zhang *et al.*, 2020). The proportion of water trapped in a material through hydrogen bonding can be determined by examining the intensity of certain peaks within the $1603\text{--}1640\text{ cm}^{-1}$ range (Chirayil *et al.*, 2014; Chandra *et al.*, 2016). The untreated vine exhibited greater peak intensity, indicating more

carboxylate groups and bound water molecules. Conversely, the bleached and acid-hydrolysed vine had fewer groups and molecules, making them more desirable as packaging materials (Chirayil *et al.*, 2014).

The degree of H-bonding in packaging materials can be inferred from key bands in the $1316\text{--}1370\text{ cm}^{-1}$, 2900 cm^{-1} , and $3300\text{--}3400\text{ cm}^{-1}$ regions (Fig. 2b) as these are linked to H-bonding (Doh *et al.*, 2020). The $1368\text{--}1370\text{ cm}^{-1}$ peaks indicate the symmetric stretching of the C-OH bonds, whereas $1316\text{--}1317\text{ cm}^{-1}$ indicate the asymmetric C-H deformations, and the 1335 cm^{-1} peak indicates the O-H bending in crystalline cellulose (Luzi *et al.*, 2014). The untreated vine spectra showed decreased intensity or absence of these bands, indicating lower H-bonding than the treated samples. This suggests that the treated samples have a more crystalline structure. The bands at $\sim 2900\text{ cm}^{-1}$ became progressively more intense and acute from the untreated to the treated samples, indicating increased inter and intramolecular H-bonding (Oh *et al.*, 2005). The broad peak centered around 3300 cm^{-1} is sensitive to the extent and type of H-bonding present (Chirayil *et al.*, 2014), with a higher wavenumber indicating more intramolecular H-bonding and a lower wavenumber indicating more intermolecular H-bonding. The comparable integrated areas under the peaks in the given region for both the bleached and acid-hydrolysed vine indicated similar levels of crystallinity. This observation was further confirmed by XRD analysis (Fig. 3). The untreated sample had a significantly lower area under the curve, suggesting a more amorphous nature. Thus, FTIR results showed that treatments removed noncellulosic parts from the raw vine, while XRD analysis was needed to determine CI more conclusively.

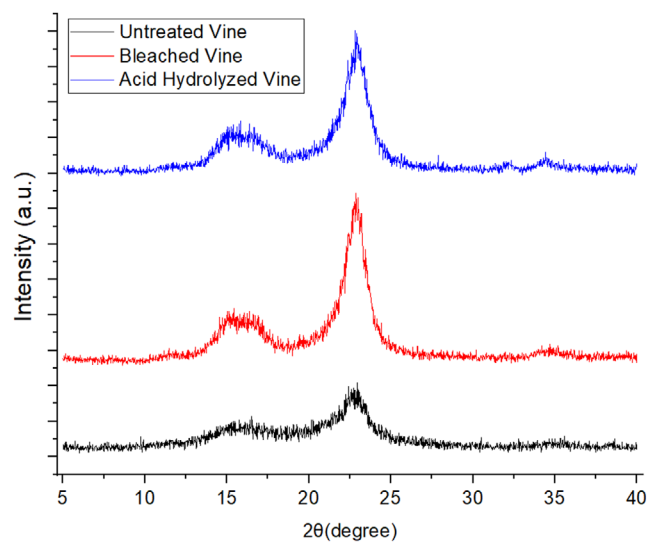


Figure 3 XRD patterns of the untreated, bleached, and acid-hydrolysed vine samples.

XRD analysis

The XRD analysis depicted in Fig. 3 and the crystallinity index (CI) values presented in Table S2 indicate the presence of peaks at $2\theta = 15\text{--}16^\circ$, $22\text{--}23^\circ$, and $34\text{--}35^\circ$, which are characteristic of the cellulose I structure (Chirayil *et al.*, 2014; Sung *et al.*, 2017; Doh *et al.*, 2020). The peak at $2\theta = 22\text{--}23^\circ$ represents the distance between the stacked sheets of adjacent cellulose chains, held together by hydrogen bonding (Luzi *et al.*, 2014). The peaks at $2\theta = 15\text{--}16^\circ$ and $2\theta = 34\text{--}35^\circ$ are indicative of the orientation of chains along the fibre length (Luzi *et al.*, 2014). The XRD patterns and reported CI values indicate that both the bleached vine and acid-hydrolysed vine have equal crystallinity, possibly due to the combined effect of the strong oxidising and subsequent hydrolytic agents used (Huang *et al.*, 2022). It is possible that acetic acid, used in the hydrolysis step, had already started to hydrolyse the glycosidic bonds of hemicellulose and amorphous cellulose before H_2SO_4 hydrolysis was conducted, which could account for the observed decrease in crystallinity index values from bleached (90.8%) to the acid hydrolysed vine (90.0%). If the amorphous fractions were already degraded by acetic acid, the H_2SO_4 hydrolysis would have only reduced particle size and crystallinity by breaking down the crystalline parts of cellulose.

The findings in Table S2 corroborate the results from the FTIR analysis, indicating that the non-cellulosic and amorphous components decreased progressively with each treatment step. The largest reduction was observed in the bleaching step, which contributed to an 11.4% increase in CI. This increase in CI upon treatment is consistent with previous studies (Sung *et al.*, 2017). Notably, the acid-hydrolysed vine had a CI of 90.0%, which is higher than the CI of sargassum seaweed (85.1%) and pure cellulose (41.4–60%) reported in other studies (Sung *et al.*, 2017; Doh *et al.*, 2020). However, the CI for acid-hydrolysed kudzu vine was not as high as the impressive CI of kombu seaweed CNCs (98.9%) isolated by Doh *et al.* (2020) using similar methods. Nevertheless, a CI of 90% is very close to perfectly crystalline cellulose and is above the 80% threshold needed to self-assemble into a chiral nematic phase with improved optical and mechanical properties. It should be noted that purely crystalline CNCs may not be ideal for packaging applications since some degree of amorphousness is beneficial in achieving desired viscoelastic and rheological properties.

Thermal analysis

The thermal degradation profiles showed some weight loss between 25 and 125 °C (Fig. 4), which was mostly caused by the evaporation of bound water

(Mondragon *et al.*, 2014; Doh *et al.*, 2020). The figure shows untreated vines displayed more weight loss, followed by bleached and acid-hydrolysed vines. The acid-hydrolysed vines lost $\sim 1.5\%$, the bleached vines lost $\sim 4\%$, and the untreated vine lost ($\sim 5.5\%$) at 25–125 °C. It is clear that more thorough treatment yields less bound water. The more crystalline cellulose in the plant cell wall is surrounded by the amorphous lignin, which serves as a barrier to keep the cellulose-bound water in place (Khalil *et al.*, 2015; Fu *et al.*, 2021). After lignin removal with successive treatment, the cellulose retained less water, and the water that was bound to the hydrophilic cellulose could quickly evaporate. Additionally, more treatment results in higher crystallinity, and the more crystalline samples were less likely to bind to and entrap water because of their more closely packed fractions (Mondragon *et al.*, 2014). Further, the bleached vine (90.8% CI) had a higher weight loss than the acid-hydrolysed sample (90.0% CI), possibly due to higher water-entrapping lignin contents. Furthermore, relatively less weight loss of the acid-hydrolysed vines could be explained by (1) the acid-hydrolysis step could serve as an intrinsically dehydrating mechanism and (2) replacement of hydroxyl groups with sulfate and acetyl groups in the amorphous fraction of cellulose led to lower H-bonding and overall higher hydrophobicity (Mandal & Chakrabarty, 2011).

Hemicellulose decomposes exothermically between 220 and 315 °C with maximum degradation around 260–270 °C (Yang *et al.*, 2007). Significant increases in derivative weight were seen in this range in the vine that had not been treated or bleached (Fig. 4b). The depolymerisation and bleaching procedure largely eliminated hemicellulose. The higher derivative weight (Fig. 4b) indicates that some hemicellulose most likely remained in the bleached sample. However, the majority of these remains were destroyed during acid hydrolysis (Doh *et al.*, 2020). As a result, near the maximum degradation temperature, acid-hydrolysed vines were substantially less impacted.

Pectin underwent exothermic pyrolysis in a reasonably broad temperature range of 200–400 °C (Mondragon *et al.*, 2014). In order to effectively remove non-cellulosic fractions while preserving cellulose, bleaching with KOH is recommended. As shown in Fig. 4a, the bleached and acid-hydrolysed materials' TGA curves remained above those of the untreated samples, which could be due to the increased pectin in the depolymerised and untreated samples. The higher weight shift in the untreated sample can be attributed to the pectin in these samples burning off. The significant DTG (Fig. 4b) for the untreated vine elevated between 180 and 220 °C. Less obviously than in the acid-hydrolysed vine samples, the bleached vine shows higher rates of pectin and hemicellulose burning off. This lower DTG rate at

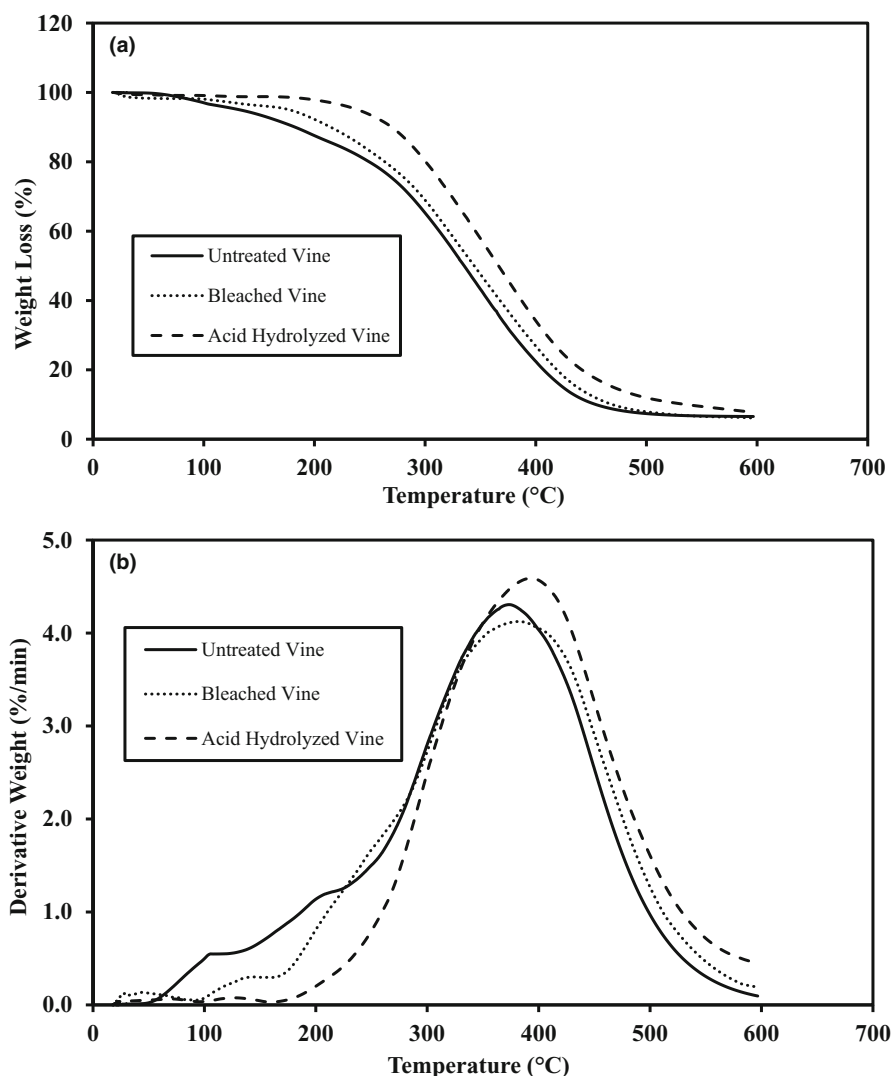


Figure 4 TGA (a) and DTG (b) curves of the kudzu vine after no treatment, bleaching, and acid hydrolysis.

180–220 °C for the acid-hydrolysed vine could be explained by the trace amounts of hemicellulose and pectin after the bleaching step (Mondragon *et al.*, 2014).

Lignin, owing to its varied composition and aromatic structure, has the broadest range of thermal degradation spanning from 150 to 900 °C. The decomposition process is exothermic until around 550 °C and then switches to an endothermic degradation process until 900 °C. Consequently, determining the exact point of maximum lignin degradation is challenging due to its extensive range. Nevertheless, samples with higher concentrations of lignin will present a wider thermal degradation profile, which is attributed to the difficulty in decomposing the diverse branches and aromatic rings present in lignin (Mandal &

Chakrabarty, 2011). This effect is reflected in the broader DTG curves of the untreated and bleached vine samples compared to the narrow acid hydrolysed sample (as depicted in Fig. 4b). It follows that the untreated and bleached vine samples have significantly more lignin than the acid-hydrolysed sample. Therefore, based on this finding, the bleached sample may contain more lignin than the acid-hydrolysed sample. Notably, the DTG curve of the bleached vine sample exhibited a sharp increase in the 150–250 °C range, which is where significant lignin degradation occurs (Yang *et al.*, 2007).

Cellulose is known to have a more uniform and less complex structure than lignin, pectin, and hemicellulose, making it more rigid, dense, and thermally stable. As a result, the thermal degradation onset occurs at a

higher temperature. Conversely, the presence of impurities such as lignin, pectin, and hemicellulose in the less treated samples may have contributed to a slower onset of thermal degradation as they lack the inherent crystallinity of cellulose and have more accessible active sites for degradation (Lee *et al.*, 2018; Doh *et al.*, 2020). Cellulose has the narrowest thermal degradation window of 315–400 °C and degrades endothermically at below 500 °C due to different mechanisms during heating compared to the non-cellulosic components (Yang *et al.*, 2007). The acid-hydrolysed sample had a larger residue, which could be attributed to higher crystallinity and more severe treatment (Mondragon *et al.*, 2014; Doh *et al.*, 2020). The more severely treated samples underwent more chemical reactions and had more molecules grafted onto the parent chain, such as acetyl and sulfate groups. The sulfated groups of the acid-hydrolysed sample also gave the structure flame resistance and accounted for its low charring rate and larger residue (Mandal & Chakrabarty, 2011). However, the presence of sulfate groups hindered thermal stability by acting as initiators for thermal decomposition and creating more interchain space. It is widely understood that treating with H₂SO₄ reduces thermal stability (Doh *et al.*, 2020).

The acid-hydrolysed sample exhibits a slower onset of thermal degradation than pure cellulose, which is likely attributed to the enhanced thermal conductivity of CNCs. The decrease in photon scattering of CNCs, compared to cellulose molecules, results in a less efficient photon pathway and poorer heat conductivity. As a result, the increased thermal conductivity of the CNCs enables more efficient and quicker thermal degradation than pure cellulose (Yang *et al.*, 2007). Nonetheless, it was observed that successive treatment led to an increase in the onset temperature, and the peaks of the DTG curve shifted to the right. This finding is believed to result from the elimination of amorphous fractions with successive treatment, enabling the reorganisation and denser packing of crystalline cellulose fractions and increased heat stability.

Conclusions

Kudzu vine is an intriguing source for the extraction of nanocellulose, specifically cellulose nanocrystals (CNCs). The CNCs extracted from kudzu possess a high aspect ratio (6.43) and length (285.01 nm), suggesting their potential suitability for various applications, including composites. Although the CNCs have a relatively large diameter, this may restrict their use in some applications. As a result, further research into higher concentrations of sulfuric acid (>51%) for CNC extraction is necessary to enhance the yield and crystallinity index. It is also important to consider the

environmental impact of using hazardous chemicals during the extraction process. If the process becomes more commercially prevalent, it may lead to negative environmental impacts. Therefore, developing sustainable extraction methods, such as enzymatic hydrolysis or steam explosion, should be a priority. Overall, the kudzu vine presents a promising avenue for the extraction of nanocellulose. However, further investigation and development of sustainable extraction methods are necessary to ensure the viability and environmental sustainability of this process.

Conflict of interest

The authors declare no conflict of interest.

Ethical approval

Ethical approval was not required for this research.

Author contributions

Duncan O. Darby: Writing – review and editing (equal). **William S. Bridges:** Writing – review and editing (equal). **Hansol Doh:** Resources (equal); supervision (equal); writing – review and editing (equal). **Reid Love:** Conceptualization (equal); data curation (equal); methodology (equal); resources (equal); writing – original draft (equal). **William Whiteside:** Conceptualization (equal); funding acquisition (equal); writing – review and editing (equal). **Sneh Punia Bangar:** Formal analysis (equal); writing – original draft (equal); writing – review and editing (equal).

Data availability statement

Data openly available in a public repository that issues datasets with DOIs

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Table S1. The dimensions of kudzu vine CNCs measured and statistically analysed *versus* the seaweed CNCs (Doh et al., 2020).

Table S2. The calculated crystallinity indexes of the kudzu vine after different stages of treatment compared to the Doh et al. (2020) seaweed samples (Segal et al., 1959).