

Preparation and Properties Research of Cellulose Nanocrystals from Flax Straw

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Abstract

This study evaluated flax straw as a renewable feedstock for cellulose nanocrystals (CNCs) prepared by alkali treatment, bleaching, and ultrasonic-assisted acid hydrolysis. Selected sulfuric-, phosphoric-, and hydrochloric-acid conditions and hydrolysis durations were compared to assess their effects on morphology, crystalline structure, chemical characteristics, thermal behavior, and dispersion performance. Transmission electron microscopy showed predominantly rod- or needle-like particles with average lengths of 200–300 nm and widths of 20–27 nm. Fourier-transform infrared spectra indicated a substantial reduction in lignin- and hemicellulose-related absorption bands while preserving the β -1,4-linked cellulose framework. X-ray diffraction confirmed retention of the cellulose I β allomorph, and the empirical Segal crystallinity index increased from 57.54% for untreated flax straw and 78.00% for cellulose microfibrils to 81.62–85.71% for the examined CNC samples. Among the conditions evaluated, CNC-S30 exhibited the highest measured crystallinity index (85.71%) and the highest apparent thermal-degradation onset, at approximately 300°C. The hydrolysis condition also influenced particle dimensions, surface charge, and dispersion behavior, indicating that the resulting CNC properties were governed by the combined effects of acid chemistry and treatment duration rather than crystallinity alone. These findings demonstrate the feasibility of converting flax straw into nanocellulose and provide a basis for further process optimization and application-oriented development of flax-straw-derived CNCs in composite and functional materials.

Introduction

Flax, widely cultivated in the northwest of China, has long been mainly used as an oil crop, with its seeds for oil extraction and the straw often burned, leading to a serious waste of natural fiber resources. Flax fiber, derived from the phloem of the straw after degumming, has a relatively short single fiber length (17–26 mm), but its cellulose content is as high as 65%–75%, second only to cotton among plant fibers. This characteristic makes it an ideal raw material for preparing cellulose nanocrystals (CNC). Recycling flax straw for CNC production can not only increase the value of agricultural waste, make up for the supply gap of cellulose raw materials, but also has the advantages of wide availability, renewability and low cost; at the same time, this utilization method helps reduce greenhouse gas emissions and landfill space occupation during the treatment of biomass waste, having a positive impact on the environment.

Natural cellulose is composed of crystalline and amorphous regions. To obtain high-purity and high-quality CNC, it is necessary to remove non-cellulose components and the amorphous region through processing. The preparation usually includes two steps: pretreatment and nanoscale dispersion. Mechanical methods, as a common approach, release nanofibrils by breaking hydrogen bonds between fibers through external force. Since they do not destroy the amorphous region, the crystallinity of the obtained CNC is similar to that of the raw material (Seta et al. 2020). A study used high-pressure homogenization to treat microcrystalline cellulose and found that after 10–15 treatments, the diameter of the resulting cellulose nanocrystals reached 28–100 nm, but agglomeration occurred after 20 treatments (Lee et al. 2009). Another study combined ball milling with maleic acid hydrolysis to obtain

well-dispersed cellulose nanocrystals from bamboo fibers, with a yield of 24.50%(Seta et al. 2020). A further study used high-intensity ultrasound to prepare rod-shaped cellulose nanocrystals with a diameter of 10–20 nm and a length of 50–250 nm. However, as the ultrasound treatment time increased, their length and crystallinity decreased, and their dispersion was poor. Although mechanical methods have a relatively low environmental impact, their equipment costs are high, energy consumption is substantial, and the products are prone to agglomeration, which limits their independent application. Therefore, mechanical methods are often combined with other methods to improve the preparation efficiency of cellulose nanocrystals(Li et al. 2012).

Oxidative hydrolysis methods achieve C6 hydroxy group oxidation to carboxy groups through selective oxidation and subsequent mechanical action for separation and dispersion to obtain CNC. Common oxidants include TEMPO, ammonium persulfate (APS), and periodate, etc. Among them, the TEMPO-mediated oxidation system (such as TEMPO/NaBr/NaClO and TEMPO/NaClO/NaClO₂) has high selectivity and efficiency(Isogai and Zhou 2019), and can achieve C6 directional oxidation while maintaining the morphology integrity and crystallinity of CNC (Rol et al. 2019). Using this method, cellulose nanocrystals (CNC) were prepared from rice straw, exhibiting a diameter of 10–25 nm and a length of 200–400 nm, along with good dispersion and thermal stability (Zhao YJ et al. 2017). However, this method involves expensive catalysts, poses pollution risks, and faces difficulties in recovering additives, which restricts its industrial application. Alternatively, the APS oxidation method can be employed to prepare CNC bearing carboxy groups. Through a one-step hydrothermal APS oxidation process under optimized conditions, a carboxylation rate of 37.89% was achieved (Ye et al. 2018). Furthermore, CNC with high crystallinity (74.4%) and high thermal stability (initial degradation temperature of 346.7°C) were obtained from lemon seeds(Zhang et al. 2020). However, the APS method still has problems such as long reaction time and low yield, which limit its large-scale application.

Acid hydrolysis is one of the most commonly used methods for preparing CNC, which can produce ultrafine and uniformly distributed nanocrystals. Its performance is affected by factors such as acid type, concentration, reaction temperature and time (Ahn et al. 2012). Through sulfuric acid hydrolysis of office waste paper, it was found that increasing acid concentration initially increased and then decreased the crystallinity of the resulting CNC, with the highest crystallinity (74.1%) achieved at 59% sulfuric acid concentration (Lei et al. 2018). Using a phosphoric acid hydrothermal method, spindle-shaped CNC were obtained after 2 hours of hydrolysis; however, hydrolysis exceeding 4 hours led to decreased thermal stability due to excessive hydrolysis (Zhou SH et al. 2019). In another study, hydrolysis with anhydrous oxalic acid successfully grafted carboxy groups onto the CNC surface, which not only enhanced its dispersibility and thermal stability (initial degradation temperature > 300°C) but also enabled material functionalization (Jia et al. 2017). Through response surface methodology optimization of the sulfuric acid hydrolysis process for bamboo pulp, the order of factors affecting CNC yield was identified as reaction temperature > reaction time > sulfuric acid concentration(Zhuo ZF et al. 2015). Traditional sulfuric acid hydrolysis presents several problems, including environmental pollution, equipment corrosion, and poor thermal stability of CNC due to the introduction of sulfate ester groups. In contrast,

organic acid hydrolysis has advantages such as low corrosivity, less pollution, and recyclability, and can endow CNC with specific functions through surface esterification, showing good application prospects.

Enzymatic hydrolysis prepares CNC by selectively degrading the amorphous region of cellulose with cellulase and retaining the crystalline region. The process mainly includes three steps: endoglucanase first attacks the glycosidic bonds in the amorphous region; exoglucanase then acts on the reducing and non-reducing ends of the polysaccharide chain, generating disaccharides and tetrasaccharides; finally, glucosidase further degrades them into glucose (Shen et al. 2022). Compared with acid hydrolysis, this method is environmentally friendly, has no toxic by-products, can recycle glucose, and use the hydrolysis residue for fermentation (Cui et al. 2016). It was found that enzyme pretreatment combined with mechanical methods could effectively dissociate jute fibers, and only a low enzyme dosage (80 mg/kg) was needed to promote the nanization of jute pulp, reduce the number of homogenization cycles, and improve production efficiency (Aguado et al. 2022). However, enzymatic hydrolysis still faces problems such as high cost, long cycle, low efficiency, and poor dispersibility of CNC, which limits its independent application. Therefore, it is often combined with mechanical or chemical methods to optimize the preparation of CNC.

This study aimed to evaluate the feasibility of producing cellulose nanocrystals from flax straw by ultrasonic-assisted acid hydrolysis and to determine how the selected acid type and hydrolysis duration affected the morphology, crystalline structure, surface chemical characteristics, thermal behavior, and dispersion performance of the resulting materials. Untreated flax straw, cellulose microfibrils (CMF), and CNC samples prepared under the investigated conditions were characterized by scanning electron microscopy, transmission electron microscopy, Fourier transform infrared spectroscopy, X-ray diffraction, thermogravimetric analysis, and zeta-potential measurements. It was hypothesized that acid hydrolysis would preferentially remove less ordered cellulose domains while largely preserving the cellulose I β allomorph, and that differences in acid identity and hydrolysis duration would produce measurable variations in crystallinity, coherent diffraction-domain size, thermal degradation behavior, and colloidal properties. By comparing the selected preparation conditions, this work sought to clarify the structure-property relationships of flax-straw-derived CNCs and to assess the potential of flax straw as a renewable feedstock for value-added nanocellulose production.

The preparation mechanism of cellulose nanocrystals

Mechanism of acid hydrolysis

The acid hydrolysis method is one of the commonly used methods for preparing CNC. Its mechanism is shown in Fig. 1. Through the action of acid on the β -1-4-glycosidic bond between two glucose units in the cellulose chain, it is protonated to form the corresponding conjugate acid, and the C-O bond of the conjugate acid breaks to form a cyclic carbocation. Finally, water molecules react with the cyclic carbocation and release H⁺ (Kumari and Singh 2018). According to the difference in chemical properties between the amorphous region and the crystalline region in biomass, the amorphous region is easily

attacked and hydrolyzed under acid hydrolysis, while the crystalline region remains intact due to its high resistance to acid attack, and thus CNC with high crystallinity is separated.

The mechanism of ultrasonic method

High-intensity ultrasonic treatment is one of the methods for preparing CNC. In the preparation of CNC by strong ultrasonic waves, ultrasonic waves propagate through the liquid medium to form an ultrasonic field. When the energy in the field is large enough, a large number of bubbles will form in the liquid. Under the influence of strong ultrasonic waves, these bubbles will go through the rarefaction and compression stages of ultrasonic waves. During these stages, the bubbles will experience a process of growth, contraction, regrowth and recontraction. After multiple periodic vibrations, they will eventually rupture at an extremely high speed, manifested as high-temperature decomposition, dispersion and shear fragmentation, which can substantially affect the surface morphology, composition, structure and chemical reactivity of the raw materials.

Materials and equipment

Sodium hydroxide (NaOH, AR) was purchased from Tianjin Fengchuan Chemical Reagent Technology Co., Ltd. (China). Sodium chlorite (NaClO₂, AR) was purchased from Shanghai Aladdin Biochemical Technology Co., Ltd. (China). Sulfuric acid (AR), hydrochloric acid (AR), phosphoric acid (AR), and glacial acetic acid (AR) were all purchased from Tianjin Damao Chemical Reagent Factory (China). An electronic balance (PL203) was obtained from Gongyi Yuhua Instrument Co., Ltd. (China). A constant temperature magnetic stirring water bath pot (HJ-A6) was purchased from Shandong Oulaibo Instrument Co., Ltd. (China). A high-speed crusher (QE-100) was supplied by Zhejiang Yili Industry and Trade Co., Ltd. (China). A freeze dryer (Scientz-10N) was purchased from Ningbo Xinzhi Biotechnology Co., Ltd. (China). A benchtop high-speed centrifuge (H1650) was obtained from Hunan Xiangyi Laboratory Instrument Development Company (China). A CNC heating ultrasonic cleaning machine (YTQX-5200DE) was purchased from Shanghai Yekuo Technology Co., Ltd. (China). A thermal field emission scanning electron microscope (JSM-7800F) was purchased from Shimadzu Corporation (Japan).

Prepare cellulose microfibrers

According to the process shown in Fig. 2, cellulose microfibrers (CMF) and cellulose nanocrystals were prepared. The specific operation method is as follows: Take 60g of flax straw and cut it into 2–3 cm sections using a grinder. Soak the ground flax straw sections in a 4% NaOH solution at 80 °C for 6 hours, replacing the NaOH solution every 2 hours. After the alkali treatment, wash the fibers with deionized water until neutral. Then, bleach the fibers with a solution composed of acetic acid buffer (27 g NaOH and 75 mL glacial acetic acid, diluted to 1 L distilled water) and sodium chlorite solution (1.7 wt% NaClO₂) at a ratio of 1:1 at 80 °C for 6 hours, replacing the solution every 2 hours. After bleaching, wash the fibers with deionized water until neutral to obtain cellulose microfibrers (CMF), with a solid-to-liquid ratio fixed at 1/20 (g/mL). The preparation process is shown in Fig. 2.

Preparation of cellulose nanocrystals by ultrasonic-assisted acid hydrolysis.

CMF was separately added to sulfuric acid solution (64 wt%), phosphoric acid solution (64 wt%), and hydrochloric acid solution (36 wt%). For each acid system, the mixture was mechanically stirred at 50°C for either 15 or 30 min, generating six hydrolysis conditions. Subsequently, the solutions were homogenized using ultrasonication for 15 min to obtain CNC suspensions. The CNC was extracted by differential centrifugation, washed with deionized water until neutral pH, and then freeze-dried using a freeze dryer. All six experimental conditions were independently repeated three times to evaluate the reproducibility of the preparation process. Three different acids and two hydrolysis times were used to examine the effects of acid type and hydrolysis duration on CNC properties. The names of the obtained products are listed in Table 1. For subsequent detailed characterization, CNC-S30, CNC-P30, CNC-C15, and CNC-C30 were selected as representative samples. CNC-S30, CNC-P30, and CNC-C30 provided a comparison of acid type at a fixed hydrolysis time of 30 min, whereas CNC-C15 and CNC-C30 provided a direct comparison of hydrolysis duration under the same acid condition. The 30 min samples were selected for the cross-acid comparison as a common longer-duration endpoint because the extended treatment was expected to promote more extensive hydrolysis of the accessible amorphous regions than the 15 min condition. Hydrochloric acid was retained at both time points because, unlike sulfuric and phosphoric acids, it does not introduce sulfate or phosphate ester groups onto cellulose; therefore, the CNC-C15/CNC-C30 comparison more directly reflects the effect of hydrolysis time rather than differences in acid-derived surface functionalization. This selection reduced redundant characterization while preserving the two principal comparisons: acid type and hydrolysis duration. A schematic flowchart of the preparation process is shown in Fig. 2.

Table 1
Names and Preparation Methods of Each Sample

Sample	Types of acids	Hydrolysis time(min)
CNC-S15	Sulfuric acid	15
CNC-S30	Sulfuric acid	30
CNC-P15	Phosphoric acid	15
CNC-P30	Phosphoric acid	30
CNC-C15	Hydrochloric acid	15
CNC-C30	Hydrochloric acid	30
Scanning electron microscopy (SEM)		

The morphology and dimensions of untreated flax straw, CMF fibers, and CNC were examined by a JSM-7800F thermal field emission scanning electron microscope (SEM). Before imaging, the samples were attached to metal specimen holders with conductive carbon adhesive tape. All samples were sputtered with gold to improve their conductivity. SEM images were collected at an accelerating voltage of 10 kV at

different magnifications. These observations were conducted to evaluate the morphological and dimensional variations of flax straw CMF and CNC resulting from chemical treatment.

Transmission electron microscope (TEM)

Transmission electron microscopy (TEM) observations were performed on a JEM-2100(UHR) instrument to examine the morphology and dimensions of the obtained CNC. A small drop of the CNC aqueous suspension (approximately 0.01 wt%) was deposited onto a copper grid and dried at room temperature. The accelerating voltage was set to 120 kV during the observation.

Fourier-transform infrared spectroscopy (FTIR)

Fourier-transform infrared spectroscopy (FTIR) measurements were performed using a PerkinElmer Spectrum Two spectrometer. Prior to analysis, the untreated flax straw, cellulose microfibrils (CMF), and cellulose nanocrystal (CNC) samples were dried to remove residual moisture and then finely ground into homogeneous powders. Each sample was thoroughly mixed with spectroscopic-grade potassium bromide (KBr) and compressed into a transparent pellet using a hydraulic press. A blank KBr pellet was first measured as the background before sample acquisition. The FTIR spectra were subsequently recorded in transmission mode over the wavenumber range of 4000–800 cm^{-1} , with a spectral resolution of 4 cm^{-1} and 16 scans accumulated for each spectrum. The measurements were repeated for each sample under the same experimental conditions. The collected spectra were subjected to background subtraction and baseline correction using instrument software, and the spectral intensities were normalized to facilitate comparison among untreated flax straw, CMF, and the CNC samples prepared under different acid-hydrolysis conditions.

Thermogravimetric analysis (TGA)

Thermal stability was evaluated by thermogravimetric analysis (TGA) under a nitrogen atmosphere (flow rate: 50 $\text{mL}\cdot\text{min}^{-1}$) using a TGA 2 instrument. Approximately 10 mg of each sample (untreated flax straw, CMF, and CNC) was heated from 40°C to 600°C at a constant heating rate of 10°C·min⁻¹.

X-ray diffraction (XRD)

X-ray diffraction (XRD) measurements were performed using an XRD-6100 diffractometer (Shimadzu, Japan). Prior to analysis, the untreated flax straw, cellulose microfibrils (CMF), and cellulose nanocrystal (CNC) samples were dried, ground into homogeneous powders, and gently pressed to form smooth, flat specimens. The diffraction patterns were collected using Cu K α radiation ($\lambda = 1.5418 \text{ \AA}$) with a Ni filter, a CM-3121 counter monochromator, and a NaI scintillation detector. The diffractometer was operated at a tube voltage of 40 kV and a tube current of 30 mA. Fixed divergence and anti-scatter slits were used, both with a beam divergence of 1.0°, and the fixed detector slit opening was 0.3 mm. The diffraction patterns were recorded over a 2θ range of 10°–70° using a step size of 0.02° and a scan speed of 5° min⁻¹. The crystallinity calculations were performed directly from the raw diffraction intensities without

smoothing, baseline adjustment, or background correction. The relative crystallinity index (CrI) was estimated using the empirical Segal peak-intensity method according to the equation(1):

$$CrI (\%) = \frac{I_{200} - I_{valley}}{I_{200}} \times 100$$

1

where I_{200} is the maximum diffraction intensity of the 200 reflection at approximately $2\theta = 22.8^\circ$, and I_{valley} is the minimum intensity between the 1–10/110 and 200 reflections, located near $2\theta = 18.6^\circ$ and used as an empirical estimate of the amorphous contribution. The apparent crystallite size normal to the (200) crystallographic plane was estimated using the Scherrer equation, and the data were calculated according to the formula(2):

$$D_{200} = \frac{K\lambda}{\beta \cos \theta}$$

2

where K is the Scherrer shape factor (0.94), λ is the X-ray wavelength (1.5418 Å), β is the full width at half maximum (FWHM) of the 200 diffraction peak expressed in radians, and θ is the Bragg angle corresponding to one-half of the measured 2θ peak position. The calculated value represents the apparent coherent diffraction-domain size normal to the (200) crystallographic plane rather than the physical particle size of the CNCs.

Zeta potential measurement

Zeta potential measurements of the CNC samples were performed on a Malvern Zetasizer Nano ZS90 instrument using dynamic light scattering (DLS). The samples were diluted to 0.1 wt% aqueous suspensions and subjected to ultrasonication at 80°C for 10 min to achieve homogeneous dispersion. Each sample was analyzed in quadruplicate, and the mean value was calculated. These measurements were conducted to assess the colloidal stability, dispersion behavior, and the effect of surface charge on the CNC particles in aqueous media.

Results and discussion

SEM

SEM characterization was conducted on flaxseed straw and CMF made after alkaline treatment and bleaching treatment, and the images are shown in Fig. 3.

As shown in Fig. 3 (a), in flax straw, water-soluble components such as hemicellulose and lignin act as binders to bond the fibers in a directional manner. In Fig. 3 (b), CMF appears as small bundles of fibers,

and individual fibers start to separate. This is because the alkaline treatment destroys some of the alkali-unstable bonds between lignin monomers or between lignin and polysaccharides, such as ether and ester bonds. In this step, the presence of chlorine in the bleaching solution oxidizes lignin and forms hydroxy, carbonyl, and carboxy groups, thereby dissolving the residual lignin. Under the combined effect of alkaline treatment and bleaching treatment, cellulose on flax straw is stripped off, forming independent fiber bundles.

TEM

In this study, CNC was isolated from purified flax cellulose, rather than from lignin, by ultrasonic-assisted acid hydrolysis. Acid molecules preferentially penetrate the more accessible disordered regions and cleave glycosidic bonds in these regions more rapidly than in the tightly packed crystalline domains, thereby releasing shorter crystalline segments. Ultrasonication improves mass transfer and promotes the separation of fiber bundles, but excessive hydrolysis severity or sonication can further shorten the particles and may begin to damage the crystalline regions. Therefore, the final CNC morphology is jointly controlled by the native microfibril architecture of flax, the effectiveness of alkali and bleaching pretreatments, acid type and concentration, reaction temperature and time, and ultrasonic energy. The TEM morphologies are shown in Fig. 4, and the average dimensions are listed in Table 2.

As shown in Fig. 4 and Table 2, the currently available descriptive average lengths of the CNC samples are 200–300 nm and the widths are 20–27 nm. The corresponding approximate aspect ratios are 15 for CNC-S15, 10 for CNC-S30, 15 for CNC-P30, and 7.4 for CNC-C30. All samples exhibit rod-like, needle-like, or slightly spindle-like morphologies, confirming that the non-crystalline regions were substantially removed while elongated crystalline domains were retained. CNC-S30 and CNC-C30 are shorter than CNC-S15 and CNC-P30, which is consistent with more extensive cleavage at structural defects and particle ends under longer or more severe hydrolysis conditions. The greater average width of CNC-C30 and its lower aspect ratio may reflect incomplete lateral separation of crystallites or the formation of small lateral aggregates.

Table 2
Average Length and Width of Each Sample

Sample	Length(nm)	Width(nm)
CNC-S15	300	20
CNC-S30	200	20
CNC-P30	300	20
CNC-C30	200	27

The dimensions obtained in this work are comparable to those reported for sulfuric-acid-hydrolyzed cellulose I nanocrystals from eucalyptus pulp (240 +/- 52 nm in length and 15.1 +/- 1.4 nm in width) and

quinoa-straw CNCs (190.29 +/- 125.25 nm in length and 20.34 +/- 4.69 nm in width). In contrast, garlic-straw CNCs have been reported to reach approximately 480 nm in length and 6 nm in width, corresponding to a much higher aspect ratio. Thus, the moderate aspect ratios of 7.4–15 obtained here are within the range reported for several plant-derived CNCs, but the differences among feedstocks can be attributed to native microfibril dimensions, pretreatment severity, acid chemistry, hydrolysis conditions, and the TEM preparation and measurement method.

The slight agglomeration and entanglement in Fig. 4 are consistent with drying-induced hydrogen bonding between the CNC surfaces rich in hydroxy groups. Acid-derived ionic groups may improve aqueous dispersibility through electrostatic repulsion, whereas low surface charge or residual electrolytes can promote reassociation. Therefore, dispersibility should be supported by zeta-potential or surface-charge data and by reporting the sonication conditions.

X-ray diffraction (XRD)

X-ray diffraction (XRD) patterns of untreated flax straw, CMF, and CNC samples prepared by ultrasonic-assisted acid hydrolysis are shown in Fig. 5. Untreated flax straw and CMF exhibited the characteristic diffraction features of native cellulose I β . The broad feature in the 2 θ region of approximately 14.7°-16.9° was assigned to the merged 1–10/110 reflections, which may overlap as the coherent diffraction-domain size decreases, rather than to an amorphous peak. The reflections at approximately 2 θ = 22.8° and 34.9° were assigned to the 200 and 004 reflections, respectively. The persistence of these reflections at similar angular positions after alkaline treatment, bleaching, and acid hydrolysis indicates that the cellulose I β allomorph was largely retained and that no evident transformation to another cellulose polymorph occurred during the treatments.

I_{200} : maximum intensity of the 200 reflection at approximately 2 θ = 22.8°.

I_{valley} : minimum intensity between the 1–10/110 and 200 reflections at approximately 2 θ = 18.6°, used as an empirical estimate of the amorphous contribution.

$$CrI (\%) = \frac{I_{200} - I_{\text{valley}}}{I_{200}} \times 100$$

Compared with untreated flax straw, CMF and the CNC samples displayed more pronounced cellulose diffraction features and a lower diffuse background. These changes are consistent with the substantial removal of amorphous non-cellulosic constituents during alkaline treatment and bleaching and with the preferential hydrolysis of less ordered cellulose domains during CNC preparation. However, XRD alone cannot demonstrate the complete removal of lignin and hemicellulose; this interpretation should be considered together with the chemical-composition and FTIR results. The essentially unchanged positions of the 1–10/110, 200, and 004 reflections further indicate that acid hydrolysis mainly altered the relative degree of lateral order and the dimensions of the coherent diffraction domains rather than changing the cellulose crystal allomorph.

The relative crystallinity index (Crl) was estimated from the raw diffraction intensities using the empirical Segal peak-height method described in the XRD characterization section. For each pattern, I200 was taken as the maximum intensity of the 200 reflection near $2\theta = 22.8^\circ$, whereas Ivalley was taken as the minimum intensity between the 1–10/110 and 200 reflections near $2\theta = 18.6^\circ$. The Crl increased from 57.54% for untreated flax straw to 78.00% for CMF, which is consistent with the removal of a substantial fraction of non-cellulosic and less ordered components during alkaline treatment and bleaching. The Crl values and the apparent coherent diffraction-domain sizes normal to the (200) crystallographic plane are summarized in Table 3.

Table 3
Crystallinity index and apparent coherent diffraction-domain size normal to the (200) crystallographic plane for each sample

Sample	Crl(%)	D200 (Å)
Flax straw	57.54	—
CMF	78.00	46
CNC-S15	82.35	43
CNC-S30	85.71	47
CNC-P30	84.58	41
CNC-C30	81.62	41

As shown in Table 3, the Crl values of the CNC samples ranged from 81.62% to 85.71%, which were higher than that of CMF (78.00%). This increase is consistent with the preferential hydrolysis and removal of less ordered cellulose domains while the more laterally ordered domains were retained (Yu et al. 2013). CNC-S30 exhibited the highest measured Crl (85.71%) among the conditions examined, followed by CNC-P30 (84.58%), CNC-S15 (82.35%), and CNC-C30 (81.62%). The apparent coherent diffraction-domain size normal to the (200) plane was 46 Å for CMF and 43, 47, 41, and 41 Å for CNC-S15, CNC-S30, CNC-P30, and CNC-C30, respectively. These Scherrer-derived values describe the apparent size of coherently diffracting domains and are not equivalent to the cellulose unit-cell dimensions or the physical particle sizes observed by TEM. The lower values obtained for CNC-S15, CNC-P30, and CNC-C30 may reflect partial disruption or fragmentation of ordered domains during hydrolysis, whereas the slightly larger value for CNC-S30 may be associated with the preferential removal of disordered material and the retention of larger coherent domains.

FTIR

Fourier transform infrared (FTIR) spectroscopy was used to evaluate changes in the chemical structure of flax straw during alkaline treatment, bleaching, and acid hydrolysis and to compare the characteristic

functional groups of the resulting cellulose microfibers (CMF) and cellulose nanocrystals (CNCs).

As shown in Fig. 6, the FTIR spectra of untreated flax straw, CMF, and CNC samples prepared under different acid-hydrolysis conditions were compared. In Fig. 6(a), untreated flax straw exhibited absorption bands at 1736, 1514, and 1249 cm^{-1} (El Achaby et al. 2018). The band at 1736 cm^{-1} was assigned to the C = O stretching vibration of acetyl and uronic ester groups in hemicellulose and/or ester linkages associated with lignin. The band at 1514 cm^{-1} was attributed to the aromatic skeletal vibration of lignin, whereas the band at 1249 cm^{-1} was associated with C-O stretching vibrations of acetyl-containing hemicellulose and lignin-related ether structures. The marked weakening or disappearance of these absorption bands in the CMF spectrum indicated that a substantial proportion of hemicellulose and lignin was removed during the alkaline and bleaching treatments. (Kassab et al. 2019; Chen et al. 2016).

Untreated flax straw and CMF retained the principal absorption bands characteristic of cellulose. The bands at approximately 1427, 1368, and 1315 cm^{-1} were assigned to CH_2 scissoring, C-H bending, and CH_2 wagging and/or in-plane deformation of hydroxy groups, respectively. The absorption bands at approximately 1160, 1106, and 1031 cm^{-1} were associated with asymmetric C-O-C stretching, skeletal vibrations of the cellulose structure, and C-O stretching in the glucopyranose units, respectively. The band at approximately 897 cm^{-1} was attributed to C1-H deformation associated with the β -1,4-glycosidic linkages of cellulose. The retention of these characteristic cellulose absorption bands after alkaline treatment and bleaching indicated that the cellulose backbone was preserved while non-cellulosic components were substantially reduced. Therefore, the FTIR results support the successful enrichment of cellulose in the CMF sample, although they do not independently establish its absolute purity (Fortunati et al. 2013).

Figure 6 (b) presents the FTIR spectra of CNC-S15, CNC-S30, CNC-P30, and CNC-C30. The CNC samples retained the principal cellulose absorption bands observed for CMF, and no substantial shifts in the main band positions were detected among the samples. This result suggests that the β -1,4-linked cellulose backbone was largely preserved under the investigated acid-hydrolysis conditions. The absorption band near 1637–1639 cm^{-1} was mainly assigned to the H-O-H bending vibration of adsorbed water in the cellulosic samples.

Overall, the FTIR spectra indicated substantial removal of lignin- and hemicellulose-related components during CMF preparation and preservation of the characteristic cellulose framework after acid hydrolysis. Differences among the CNC spectra were primarily reflected in band intensity and weak secondary features rather than in the appearance of unequivocal new structural bands. Consequently, any relationship between the weak band near 1731 cm^{-1} and the dispersion stability or thermal behavior of the CNC samples should be presented as a tentative interpretation and supported by complementary surface-chemical characterization.

Result of TGA

After alkali treatment, bleaching, and ultrasonic-assisted acid hydrolysis, the chemical composition, degree of structural order, chain length, and surface chemistry of flax-derived cellulose were altered. The thermal behavior of raw flax straw, CMF, and the four CNC samples is compared in Fig. 7.

As shown in Fig. 7 (a-b), raw flax straw exhibits broad and overlapping mass-loss events between approximately 175 and 400°C. This behavior originates from its heterogeneous lignocellulosic composition: low-molecular-weight extractives and hemicellulose decompose relatively early, cellulose contributes the dominant mass-loss event, and lignin decomposes over a much broader temperature interval.

Below 150°C: the main mass loss is associated with the evaporation of physically adsorbed water and weakly bound moisture.

Between 150 and 240°C: degradation of low-molecular-weight extractives and the less thermally stable hemicellulose fraction may occur together with the initial dehydration of cellulose.

From 240 to 400°C: the dominant cellulose degradation takes place through depolymerization, cleavage of glycosidic bonds, dehydration, decarbonylation, and decarboxylation, while lignin degradation continues to overlap with this region.

Above 400°C: the remaining lignin-derived and carbonaceous residues undergo slower condensation and aromatization reactions. Describing these residues as fully graphitic would require additional structural evidence.

After alkali treatment and bleaching, CMF begins to decompose at approximately 250°C and shows one dominant DTG peak centered near 350°C. Alkali treatment removes a large proportion of hemicellulose, extractives, and ester-linked lignin-carbohydrate complexes, while bleaching oxidatively removes residual lignin. Consequently, the low-temperature shoulders become less evident and the degradation behavior is increasingly dominated by cellulose. The narrower and higher-temperature degradation event is therefore consistent with increased cellulose purity and a larger proportion of ordered cellulose domains.

Figure 7 (c-d) compares the TG/DTG curves of CNC-S15, CNC-S30, CNC-P30, and CNC-C30. Although each CNC sample shows one main DTG peak, their apparent onset temperatures differ considerably. Acid identity and hydrolysis time affect thermal stability through competing mechanisms. Preferential hydrolysis of accessible amorphous domains can increase the relative crystallinity, whereas chain scission decreases the degree of polymerization. At the same time, acid-derived surface groups and residual ions may catalyze dehydration and char formation. Acid concentration, reaction temperature, and hydrolysis time also change CNC dimensions and specific surface area, which can further influence heat and mass transfer during degradation.

CNC-S15 and CNC-C30 show an apparent degradation onset near 170°C, whereas CNC-P30 and especially CNC-S30 show higher thermal stability, with CNC-S30 beginning to degrade near 300°C. The

difference of approximately 130°C between the lowest and highest onset temperatures cannot be explained by crystallinity alone. For sulfuric-acid-derived CNCs, surface sulfate half-ester groups are known to catalyze dehydration, lower the onset temperature, and increase char formation; neutralization or removal of these groups can restore thermal stability toward 300°C.

Within the CNC-S series, increasing the hydrolysis time from 15 to 30 min coincides with a reduction in average particle length from 300 to 200 nm (Table 2) and an increase in the apparent thermal stability. This result suggests that the longer treatment may have removed a greater proportion of thermally labile amorphous segments.

The stability of cellulose nanocrystal (CNC) dispersions

The dispersibility and colloidal stability of CNC in aqueous and organic media are fundamental for its application in the form of dispersions. Therefore, the Zeta potential of CNC was measured using a nanoparticle size and Zeta potential analyzer (DLS).

Zeta potential is a key indicator used to describe the stability of particles in a dispersed fluid. When the absolute value of this potential increases, the repulsive effect between charged particles becomes more pronounced, thereby enhancing the stability of the dispersion system (Mahmud et al. 2019; Cheng et al. 2017).

As shown in Fig. 8 the Zeta potential values of the extracted CNC-S15, CNC-S30, CNC-P30, and CNC-C30 were measured. The Zeta potential test results indicate that during hydrolysis with sulfuric acid, hydrochloric acid, and phosphoric acid, sulfate ester groups, phosphate ester groups, and carboxylate groups were introduced onto the surfaces of CNC-S, CNC-P, and CNC-C, respectively. This imparts a negative surface charge to the extracted CNC. Consequently, the plotted mean values showed apparent differences in the absolute zeta potential among the CNC samples.

As shown in Fig. 9, the dispersion of CNC obtained under different preparation conditions in organic solvents is presented. Within the first hour, CNC obtained under different preparation conditions can be completely dispersed in organic solvents; after 12 hours of standing, CNC-P30 begins to precipitate, while no visible changes are observed in the other samples; after 24 hours of standing, CNC-P30 almost completely precipitates, and CNC-C30 begins to precipitate, while CNC-S15 and CNC-S30 show no visible changes. This is because the Zeta potential values of CNC-S15 and CNC-S30 are relatively higher, allowing for more stable dispersion. The relatively stable dispersion of CNC-S15 and CNC-S30 in organic solvents provides possibilities for their subsequent application in electrospinning.

Conclusions

Cellulose nanocrystals (CNCs) were successfully isolated from flax straw through sequential alkaline treatment, sodium chlorite bleaching, and ultrasonic-assisted acid hydrolysis, supporting the feasibility of flax straw as a renewable lignocellulosic feedstock for CNC preparation. The resulting CNCs exhibited

predominantly rod-like or needle-like morphologies, with approximate lengths of 200–300 nm and widths of 20–27 nm. FTIR analysis indicated a marked decrease in absorption bands associated with lignin- and hemicellulose-related components, while the characteristic cellulose-related bands remained evident. XRD analysis further indicated that the cellulose I β structure was largely retained after treatment. The empirical Segal crystallinity index increased from 57.54% for untreated flax straw and 78.00% for CMF to 81.62–85.71% for the investigated CNC samples, which is consistent with the preferential removal of less ordered components and domains during pretreatment and acid hydrolysis. TGA showed that the thermal-degradation behavior varied with the hydrolysis conditions. Among the investigated samples, CNC-S30 exhibited the highest measured Segal crystallinity index (85.71%) and comparatively delayed thermal degradation. However, these observations identify the best-performing sample only within the experimental conditions evaluated and should not be interpreted as establishing a universally optimal preparation condition.

Several limitations of the present study should be considered when interpreting these findings. First, the Segal method provides an empirical relative crystallinity index and does not represent an absolute crystalline fraction; therefore, the reported values are more appropriately used for comparative assessment among samples prepared and analyzed under the same conditions. Second, although FTIR revealed changes in the chemical environment of the cellulose-rich material, FTIR alone cannot unambiguously confirm the formation or quantify the abundance of specific surface functional groups, such as sulfate ester, phosphate ester, or carboxy groups. Third, the investigated hydrolysis conditions covered only a limited experimental range, and the observed differences among the CNC samples therefore cannot be generalized to other processing conditions without further validation. More extensive statistically replicated experiments and complementary surface-chemical characterization would be required to establish more robust structure–property relationships.

Despite these limitations, the present results indicate that flax straw, an underutilized agricultural residue, has potential as a raw material for producing value-added nanocellulose while largely retaining the cellulose I β structure. The study provides useful experimental information on how pretreatment and acid-hydrolysis conditions influence the morphology, relative crystallinity, and thermal behavior of flax-straw-derived CNCs. These findings contribute to the valorization of flax straw and broaden the range of lignocellulosic resources that can be considered for CNC production. Further work should focus on systematic process optimization, quantitative surface-chemistry analysis, statistically supported comparisons, scale-up assessment, and evaluation of the performance of the resulting CNCs in composite and other cellulose-based functional-material applications.

Declarations

Declaration of conflicting interests

The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

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Author Contribution

J. Chen was responsible for conceptualization, investigation, data collection and analysis, and writing the original draft. D. Shi contributed to methodology, writing—review and editing. Cao prepared Fig. 2, and together with Z. Song, contributed to software support, data visualization, and validation. All authors reviewed and approved the final version of the manuscript.

Data Availability

No new data were created or analyzed in this study.

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Figures

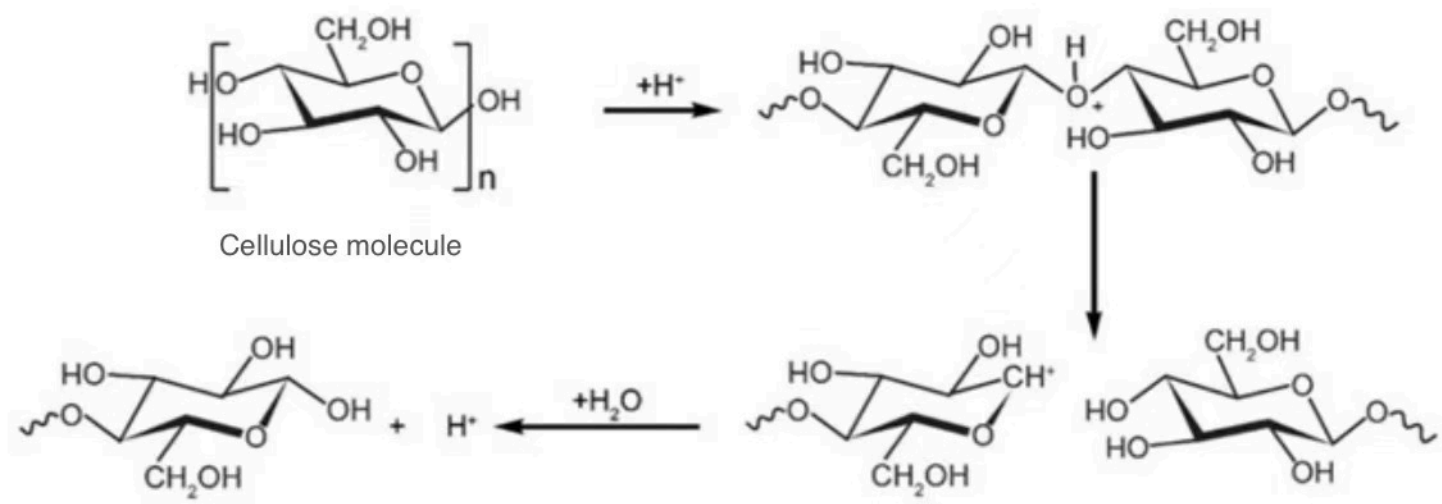


Figure 1

Fig. 2.1 Schematic Diagram of Acid Hydrolysis Mechanism

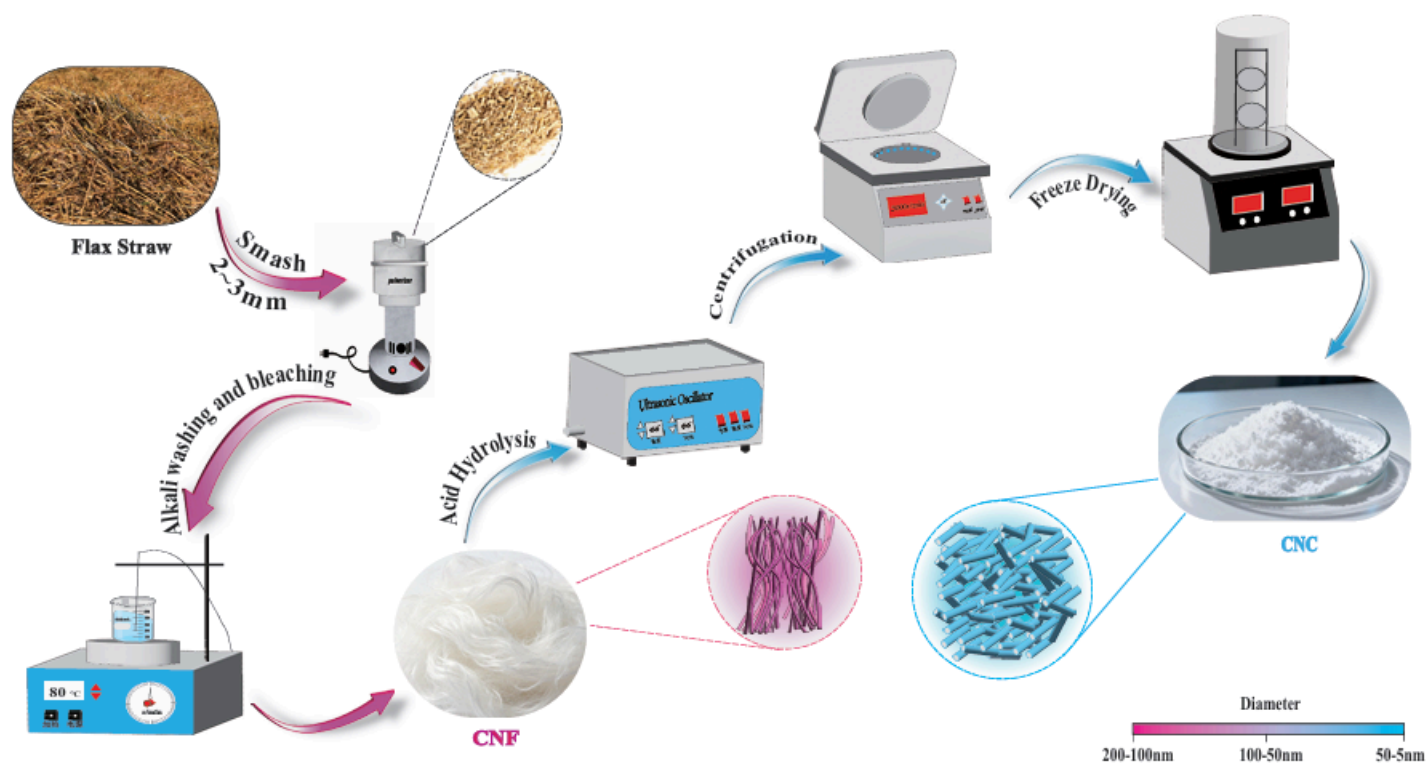


Figure 2

Fig. 3.1 Flowchart of CMF and CNC preparation process

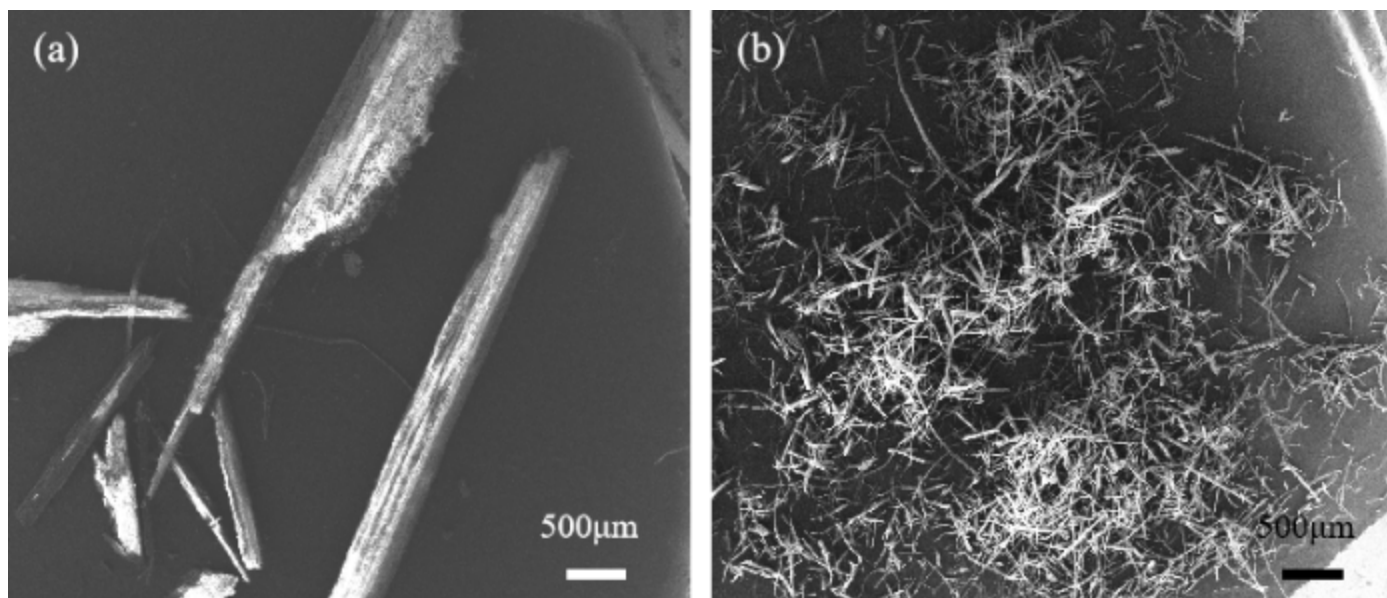


Figure 3

Fig. 4.1 SEM images of sesame (a) and CMF (b)

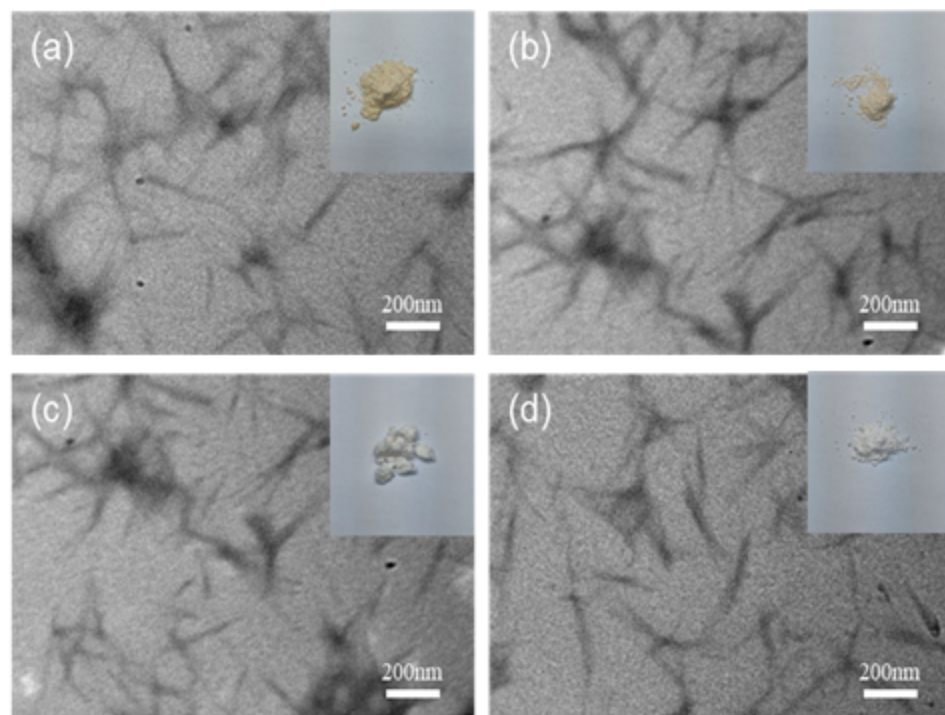


Figure 4

Fig. 4.2 TEM images of CNCs prepared by different methods (a) CNC-S15; (b) CNC-S30; (c) CNC-P30; (d) CNC-C30

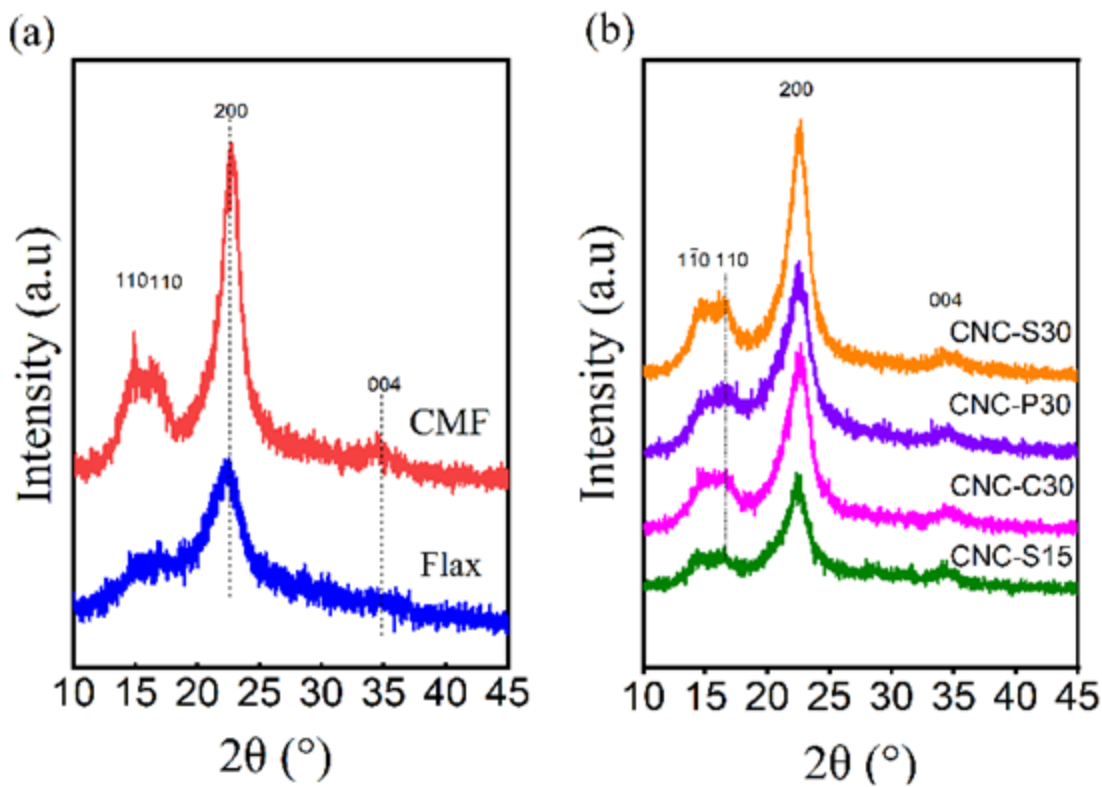


Figure 5

Fig.4.3 XRD diffraction patterns of each sample (a) flax, CMF; (b) CNC-S15, CNC-S30, CNC-P30 and CNC-C30

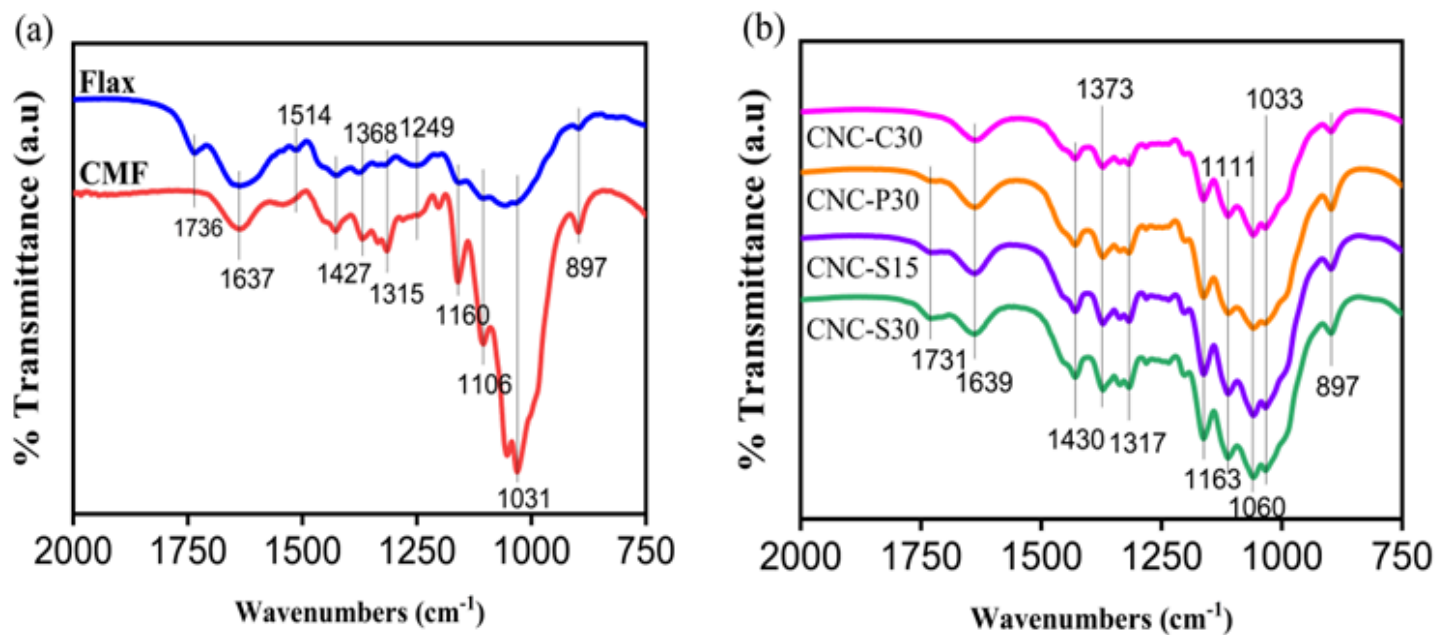


Figure 6

Fig. 4.4 Infrared spectra of each sample (a) Flax and CMF; (b) CNC-S15, CNC-S30, CNC-P30 and CNC-C30

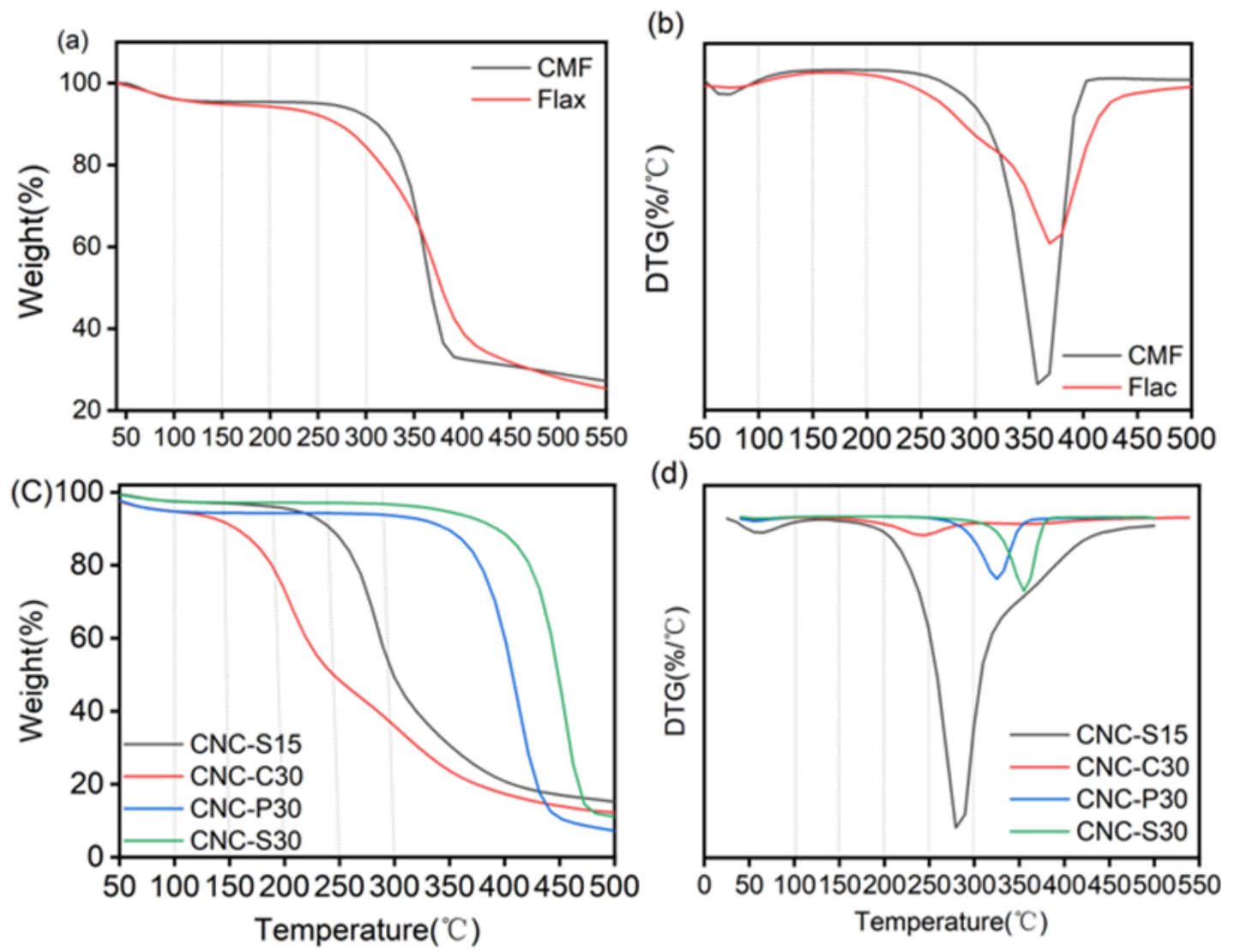


Figure 7

Fig. 4.5 (a-b) TG curves and DTG curves of flax and CMF; (c-d) TG curves and DTG curves of CNC-S15, CNC-S30, CNC-P30 and CNC-C30

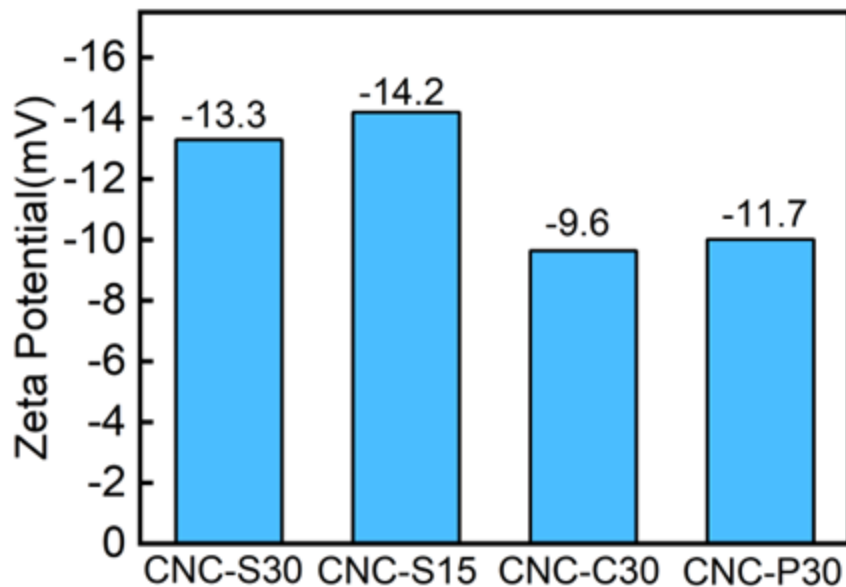


Figure 8

Fig. 4.6 The Zeta potential diagrams of CNC-S15, CNC-S30, CNC-P30, and CNC-C30

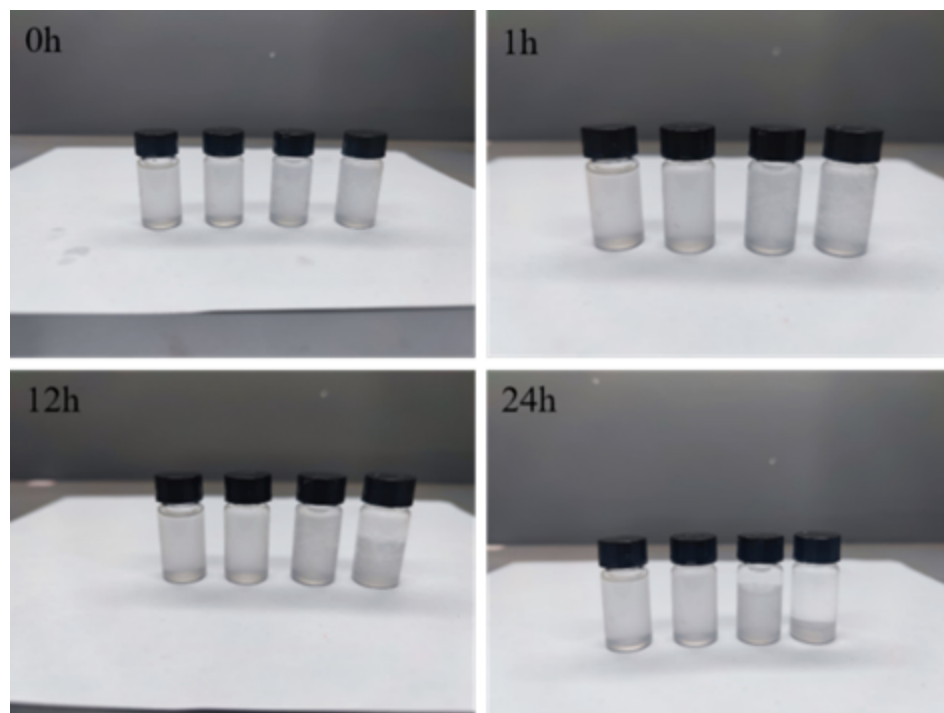


Figure 9

Fig. 4.7 depicts the dispersion of each sample in organic solvents (from left to right: CNC-S15, CNC-S30, CNC-C30, CNC-P30)