

Extraction, characterization, and fabrication of cellulose biopolymer sheets from Pistia stratiotes as a biodegradative coating material: An unique strategy for the conversion of invasive weeds into value-added products

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Abstract

Water lettuce (*Pistia stratiotes*) was used in the study to extract cellulose biopolymer. Cellulose extraction was carried out by alkali treatment followed by bleaching. Different methods for extraction of cellulose were carried out and the method which gave high yield was selected. Functional properties like bulk density, packed density, hydrated density, water retention capacity, oil retention capacity, emulsifying activity and setting volume of the extracted cellulose were studied. Quantitative analysis of cellulose was carried out by the anthrone method and the cellulose content was found to be $38.94 \pm 0.10\%$. Preliminary confirmation of cellulose biopolymer was done using the study of functional groups using FT-IR analysis. Further characterization studies like SEM, XRD, DSC and TGA were done to understand the molecular architecture and purity of cellulose extracted. Fabrication of cellulose sheets were carried out using starch as plasticizer. Biodegradation studies were carried out in garden soil for four weeks and a high degradation rate of $78.22 \pm 0.71\%$ was observed in the fourth week of soil burial.

1. Introduction

Aquatic plants are essential in maintaining the ecosystem balance in the environment. The threats from aquatic plants arise when their growth in the water system becomes too dense. The exotic varieties tend to overgrow and invade areas which are the breeding waters of many fishes. These plants also interfere with many recreational activities such as swimming and fishing. The overgrowth of these plants can be a result of high levels of nutrient influx contributed by the runoff of agricultural wastes and sewage-disposal drainage, which significantly escalates the growth (Keller et al., 2018). When non-native plants are introduced into water bodies, they perish due to the absence of their native natural predators. The aquatic exotic plants which are causing the main problems in water bodies are water hyacinth, water lettuce and water moss (Pejchar & Mooney, 2009). These non-native plants, which intrude into the ecosystem and often pose severe threats to the indigenous fauna and flora (College of Marine Science, 2000). Multiple strategies were developed for aquatic weed management; especially composting and using them for biogas production through anaerobic generation. These weeds are important sources of polymeric substances like cellulose that have a high demand in the global market due to its widespread application in various sectors. Utilization of water weeds for biopolymer production is in its dawn and requires more focus in the years ahead.

Cellulose is one of the primary components, which is primarily present in the cell walls of plants, photosynthetic organisms, and various microorganisms. It is a crystalline polysaccharide that constitutes a linear chain of repetitive units of glucose interconnected through $\beta(1 \rightarrow 4)$ linkages (King, 1982; Tony, 2008). However, various pretreatment processes viz., physical, chemical, biological, or the integrated process can significantly reduce the crystallinity, which enhances the production of monomer, glucose (Krumm et al., 2016).

Owing to the attractive physical and chemical properties of biopolymers possesses innumerable applications in different industries such as food, paper, pharmaceuticals, etc (Lima et al., 2004; Stephen &

Phillips, 2016; Dongowski, 1997). Other than these applications cellulose biomass is also used as a potential feedstock for ethanol production globally (Sassner et al., 2008; Sun et al., 2005). The extraction procedure of cellulose from the plant biomass proceeds through drying and powdering to maximize the surface area for better chemical reaction. The crude fibres can be broken down by milling, grinding or cutting in mill (Deepa et al., 2011; Rosa et al., 2010; Sundari & Ramesh, 2012). In the next step the cementing molecules (lignin and hemicellulose) should be removed, as the presence of these molecules can interfere with the crystallinity of the cellulose fibres (Faruk et al., 2012). Hemicellulose and lignin tend to reduce the crystallinity ratio, thermal stability and other mechanical properties (Bao & Li, 2011). The purification techniques remove the hemicellulose and lignin, as well as prevents moisture absorption, and improves the surface properties of the extracted fiber (Kalia et al., 2011). Alkali bleaching treatment is the most preferred purification method, among the different methods. This method is popular because of the easy availability of KOH/NaOH, sodium chlorite and acetic acid. Alkali treatment and bleaching removes lignin and hemicellulose, and even divides the fibres into smaller parts, with almost similar dimensions and even microfibrils (Karimi et al., 2014; Nurain et al., 2012; Sonia et al., 2013). An important modification happening during the alkali treatment is the dissociation of OH bondings in the structure of the fibre. The reaction happens by ionizing the hydroxyl groups to become alkoxide ($\text{Fibre-OH} + \text{NaOH} \rightarrow \text{Fibre-O-Na}^+ (\text{alkoxide}) + \text{H}_2\text{O}$) (Majeed et al., 2013), hence separate the inter-fibrillar regions from the cellulose fibers. The removal of the hemicellulose and other non-cellulosic materials, including pigments will create void within the fibre, which results in swelling fine structure, which changes the physical structures, morphology, dimensions and mechanical properties (Kalia et al., 2011; Majeed et al., 2013).

Alkali treatment will remove all the main hemicellulosic impurities, but leaves behind some lignin materials. These lignocellulosic contents are the hardest ones to separate from the crude cellulose (Jonoobi et al., 2009). Bleaching is done to remove these lignin components, bleaching of cellulose is also called as delignification (Shi et al., 2011). Most common of the bleaching treatment is boiling the alkali treated fibres with sodium chlorite under acidic conditions (Sundari & Ramesh, 2012). The acidic condition is created by the acetate buffer which contains glacial acetic acid. In the acidic condition sodium chlorite gets broken down into chlorine dioxide (ClO_2), which liberates chlorine gas during the bleaching. The ClO_2 can oxidize the aromatic rings in lignin molecules and thereby release the lignin components from the fibre into the liquid (Shin et al., 2012; Zainuddin et al., 2013). The chlorine dioxide will also oxidize any hemicellulose, pectin which were not removed by the alkali treatment. Other common bleaching agents are hydrogen peroxide and sodium hypochlorite (Hon-Meng et al., 2015). After the bleaching step, the obtained cellulose will be almost pure, but we generally employ acid hydrolysis, for obtaining high purity cellulose (Alemdar & Sain, 2008).

This research work explores the possibility of integrating weed management and biopolymer production following chemical extraction methods. An attempt was made to compare the effectiveness of an eco-friendly method employing soap nut powder as an alternative to chemical bleaching agent in cellulose extraction. The work further reports a simple method for development of biodegradable cellulose sheets followed by evaluation of their biodegradation rate using standard soil burial method.

2. Materials And Methods

2.1 Substrate collection

The water lettuce plants used in this study were collected from Meenachil river, Kottayam, Kerala, India. The collected plants (except the roots) were sun dried for 3 days (Fig:1). It was then dried at 60°C in a hot air oven for 3 days to remove the water content completely and to achieve a uniform texture. The dried plants were powdered and stored for further extraction works.

2.2 Process optimization for extraction of cellulose from *Pistia stratiotes*

2.2.1 Method 1

Powdered water lettuce was subjected to alkali treatment using 10% NaOH as suggested by Bhattacharya et al. (2008) with minor modification. 10 g of water lettuce powder was treated with 150 mL of 10% NaOH at 70 °C for 2 h. The alkali treatment was repeated thrice, after which the sample was filtered and sequentially washed with tap water and then with distilled water till the pH reached neutrality. The alkali treated substrate was then bleached with 200 mL of 2% (w/v) sodium chlorite and 2%(v/v) glacial acetic acid at 80 °C for 2 h. This step was repeated 2 times. After the bleaching procedure, pH was neutralized using distilled water and the bleached fibres were retained at 60 °C till the fibres were completely dry (Bhattacharya et al., 2008; Wang et al., 2019).

2.2.2 Method 2

In this method 10 g of water lettuce powder was initially treated using 150 mL of 4% NaOH for 4 h at 80 °C, this step was repeated three times. The mixture obtained after alkali treatment was excessively washed using distilled water till the pH turned neutral. The obtained powder was then bleached using 50 mL of 4% sodium hypochlorite at 30 °C for 3h. This step was repeated twice. After bleaching pH was neutralized using distilled water and the bleached fibres were retained at 60 °C in a hot-air oven till the fibres were completely dry (Adinugraha et al., 2005; Wu et al., 2019).

2.2.3 Method 3

The alkali treatment was performed on the water lettuce powder (10g) using 150 mL 4% NaOH for 2 h at 60 °C; alkali treatment was done thrice and the residue obtained after filtration was washed with distilled water till the pH turned neutral. Bleaching was performed by treating the alkali treated sample with 200 mL solution of 4% hydrogen peroxide in 4%NaOH for 2 h at 60 °C. After bleaching the sample was washed with distilled water till the pH becomes 7 (J. Sun, 2004; Ventura-Cruz & Tecante, 2019) and incubated at 60 °C (hot air oven) till the fibres are completely dry.

2.2.4 Method 4

For the eco-friendly bleaching method, 15g of powdered water lettuce was steam exploded for effective bleaching using 400 mL of soapnut solution (4%) and pretreated at 121 °C at 20lbs for 1h. After bleaching, the bleached fibres were washed to remove all the soapnut residues. The bleached fibres were then heated with 90 mL of acetic acid (80%) and 10 mL of nitric acid (45%) and incubated for 45 min at 100 °C (Reshmy et al., 2020; R. Sun & Tomkinson, 2005). After which the fibres were cleaned to remove acid residues with distilled water. These fibres were then treated with 100 mL of 20% acetic acid, 5 mL sulphuric acid and 2g NaCl solution at 80–90 °C for 30 min. Again this treated residue is washed with distilled water until the pH turns neutral, then kept at 60 °C in a hot air oven till the fibres are completely dry (Reshmy et al., 2021).

Based on the yield of cellulose obtained from each of the methods, the method that gave the highest cellulose yield was fixed for further characterization studies.

2.3 Quantitative analysis of cellulose

The qualitative analysis of cellulose was performed by anthrone test. The test was carried out after the cellulose was treated with solution of 15 mL of acetic acid [80% (v/v)] and 1.5 mL of nitric acid for the removal of impurities. The purified cellulose was then dissolved in 80% sulphuric acid and then diluted for the anthrone assay (Qian et al., 2020; Viles & Silverman, 1949).

2.4 Analysis of functional properties of the extracted cellulose

The functional property of the extracted cellulose was tested by determining the bulk density, packed density, hydrated density, water retention capacity, oil retention capacity, emulsifying activity and settling volume (Gichuki et al., 2020; R. Singanusong et al., 2014).

2.4.1 Bulk Density

The extracted cellulose after complete drying (0.24g) was taken in a 10 mL measuring cylinder and shaken slightly, then the volume of the sample was recorded (Gichuki et al., 2020; Prakongpan et al., 2002). The bulk density was determined as stated below

$$\text{Bulk density (g / mL)} = \left(\frac{\text{amount of sample (g)}}{\text{volume of sample (mL)}} \right)$$

1

2.4.2 Packed Density

In a 5 mL graduated syringe 0.24g of dried cellulose was taken and the piston was pushed to apply pressure on the cellulose. The piston was pressed till the cellulose sample would not reduce further in volume (Gichuki et al., 2020; Prakongpan et al., 2002). The packed density was calculated as follows:

$$\text{Packed density (g / ml)} = \left(\frac{\text{amount of sample (g)}}{\text{least volume of sample (mL)}} \right)$$

2

2.4.3 Hydrated Density

In a 10 mL measuring cylinder, 5 mL distilled water was added and then to this cylinder, 0.241g of dry cellulose was also added. The volume of water displaced after adding cellulose was measured (Prakongpan et al., 2002). The hydrated density is found out by the equation:

$$\text{Hydrated density (g / mL)} = \left(\frac{\text{sample (g)}}{\text{mL of water displaced}} \right)$$

3

2.4.4 Water Retention Capacity (WRC) and

In a 50 mL centrifuge tube 2g of the extracted cellulose was taken and mixed well with 30 mL of distilled water. The resulting slurry was left undisturbed for 10 min, then was centrifuged at 2000 rpm for 15 min. After centrifugation the pellet was collected by draining out the supernatant and was weighed as the wet sample weight (Singanusong et al., 2014). The WRC was calculated by the formula:

$$\text{Water Retention Capacity (g / g)} = \left(\frac{\text{weight of water (g)}}{\text{weight of dry sample (g)}} \right)$$

4

2.4.5 Oil Retention Capacity (ORC)

The ORC was also calculated by following a similar procedure to WRC, except that instead of water palm oil was used. The ORC was calculated by the formula:

$$\text{Oil Retention Capacity (g / g)} = \left(\frac{\text{weight of oil (g)}}{\text{weight of dry sample (g)}} \right)$$

5

2.4.6 Emulsifying Activity (EA)

For calculating EA, 7g of the extracted cellulose was added into 100 mL water and subsequently to 100 mL of soybean was added. This mixture was homogenized at 1000 rpm for 1 min. The emulsion obtained was centrifuged at 1300rpm for 5 min (Singanusong et al., 2014; Prakongpan et al., 2002). The EA was calculated by the following equation:

$$\text{EmulsifyingActivity} = \left(\frac{\text{Height of emulsifier level (cm)}}{\text{Height of whole layer (cm)}} \right) \times 100$$

6

2.4.7 Settling volume (SV)

SV was measured by mixing 1g of the extracted cellulose in 70 mL of distilled water, in a 100 mL screw cap bottle. To saturate the cellulose sample and to remove the excess gas trapped in the mixture, the bottle was subjected to 30 min of ultrasonic treatment. For degassing, the mixture was vacuum sucked for 30 min, after which it was kept in the cold room for 24 h. This will allow water to penetrate to the interstices of the cellulose sample. The mixture was then transferred into 100 mL volumetric flasks and were quantitatively measured and made upto 100 mL by adding distilled water. The settling volume is the value given by the sample residue layer when the mixture was left undisturbed at room temperature for 24 h (Singanusong et al., 2014).

2.5. Characterization of extracted cellulose

The functional and structural characterization of cellulose biopolymer extracted from water lettuce by different analytical techniques. The functional groups of the biopolymer were examined between the IR range frequency of $4000 - 400 \text{ cm}^{-1}$ using an FTIR spectrophotometer (Shimadzu IR Spirit-L). The morphology of the extracted cellulose was determined via a scanning electron microscope (SEM, Jeol JSM 6390). The purity and crystal phase of the cellulose from water lettuce was elucidated by X-ray diffractometer analysis (Shimadzu XRD 6000). Differential scanning calorimetry (DSC) analysis of cellulose samples was exposed to a temperature range of -50°C to 400°C at a rate of $10^\circ\text{C}/\text{min}$ using a DSC instrument (Netzsch DSC 204 F1). Thermogravimetric analysis (TGA) of cellulose was done using a Perkin Elmer STA 6000 from room temperature (25°C) to 600°C as the heating range.

2.6. Fabrication of cellulose sheets

Cellulose sheet was prepared using distilled water and plasticizer. Approximately 2.5 g of extracted cellulose was added in 100 mL distilled water and stirred for 6 h. This was then homogenized at 7400 rpm for 20 min and was kept in a cold room at 4°C overnight. The sample was then homogenized again under the same conditions and incubated at 60°C in an aluminum tray (Reshmy et al., 2020).

Plasticizers are added during the second homogenization step; the plasticizer used was 1% starch and casting was done (Reshmy et al., 2021).

2.7. Biodegradation study of cellulose sheets

Pre-weighed cellulose sheets (CS) prepared from water lettuce were embedded inside a grid case (non-corroding) and buried into a plastic cartridge containing 100 g of soil samples (garden soil). Pre weighed cellulose acetate filter paper sheets (FP) and cellophane tapes (CT) prepared in the same way were buried in garden soil to serve as comparative standard. Moisture content of the soil was maintained at 60% by addition of water regularly. All the containers were incubated at room temperature ($30 \pm 5^\circ\text{C}$) for a period

of 28 days. The weight of the CS was monitored at regular intervals (7 days) by washing the residue with distilled water and drying at 60 °C prior to weighing. The percentage degradation as a function of the weight loss in relation with the number of days was determined as per the equation suggested by Kim et al. (2000).

$$\% \text{Degradation} = \left(\frac{CS_{Initialweight} - CS_{Finalweight}}{CS_{Initialweight}} \right) \times 100$$

7

2.9. Statistical analysis

Each experiment was performed in triplicate and presented along with the standard error data. The significant difference between CS, FP, and CT for the biodegradation studies was determined using Minitab 17.1.10 version ($p < 0.05$).

3. Results And Discussions

3.1. Extraction of cellulose

Four different methods for cellulose extraction were done and compared for cellulose yield (Table 1). Method 1 gave the highest yield of $38.92 \pm 0.38\%$ (Fig. 2(a,b)) and was selected for further studies.

Table 1
Comparison of different cellulose extraction methods

Method	Bleaching agent used	% Yield of Cellulose (w/w)
Method 1	2% Sodium chlorite and 2% Glacial acetic acid	38.92 ± 0.038
Method 2	4% Sodium hypochlorite	25.70 ± 0.035
Method 3	4% Hydrogen peroxide + 4% Sodium hydroxide	10.7 ± 0.0377
Method 4	4% soapnut solution	NA
*Data represents mean $\pm$ SD values performed in triplicate which is significant at $P < 0.05$.		

In method 1 alkali treatment was done using NaOH which removes all the hemicellulose contents. The components left behind are lignocellulosic contents and other impurities. Bleaching with sodium chlorite under acidic conditions will cause delignification (Sundari & Ramesh, 2012). The sodium chlorite will break down into ClO₂ under an acidic environment. ClO₂ will liberate chlorine gas during the bleaching process by oxidizing the aromatic rings of lignocellulosic materials. This reaction releases the lignin into the liquid, leaving non-soluble cellulose fibres (Shin et al., 2012; Trilokesh & Uppuluri, 2019; Zainuddin et al., 2013). Thus, the cellulose extracted from P. stratiotes using alkali and bleaching pretreatment yielded

38.94 ± 0.1%. Melikoğlu et al. (2019) reported the extraction of cellulose from apple pomace by the optimized alkali pretreatment process (10.23% NaOH, 69.82 °C, and 161.54 min) was found to be 27.96 ± 0.78%. The enhanced extraction of cellulose can also be attributed to the presence of higher content of cellulose (40.63%) in the water lettuce biomass (Sunanda et al., 1991). However, the eco-friendly method followed did not provide the expected result as the bleaching of the water lettuce powder was not proper. This may be due to different chemical constituents present in water lettuce which can reduce the mild bleaching activities of soapnut powder which is routinely used as a natural bleaching agent.

3.2. Analysis of functional properties

The functional properties of cellulose extracted from water lettuce was compared with commercial cellulose standard (Table 2). The values of bulk density packed density and hydrated density of the cellulose isolated from water lettuce were comparatively significant over commercial cellulose (CC) reported in the literature (Singanusong et al., 2014). This may be due to the smaller particle size of commercial cellulose (Prakongpan et al., 2002). In a study dealing with extraction of cellulose from pineapple cores the bulk density, packed density and hydrated density were observed to be 0.171 ± 0.003, 0.385 ± 0.013 and 1.47 ± 0.36 (g/mL) respectively (Prakongpan et al., 2002). In another study dealing with the extraction of cellulose from banana peels using NaOH and hydrogen peroxide, these bulk density (0.646 ± 0.27 g/mL), packed density (0.923 ± 0.07 g/mL) and hydrated density (2.50 ± 0.48 g/mL) reported were in close agreement with this study (Singanusong et al., 2014). The WRC of the cellulose obtained from water lettuce was observed to be higher than that of CC probably due to its larger particle size of fibers that accelerate interaction between water. The WRC value of cellulose extracted from banana peels and pineapple core was found to be 2.91 g water/g dried sample and 9.92 ± 0.1g water/g dried sample of fiber respectively. The WRC of the cellulose extracted can be reduced by reducing the particle size of the extracted cellulose for further application studies. On the contrary, cellulose with higher WRC is preferred for improving its texture especially for packaging applications. The ORC of cellulose varies with the raw material employed for extraction. In this study, the ORC of cellulose from water lettuce was significantly higher than the commercial cellulose and is in agreement with the ORC of cellulose extracted from pineapple core fiber (Prakongpan et al., 2002). The higher emulsifying activity of the extracted cellulose as compared to the CC can be clearly related to its higher ORC. The settling volume and EA data obtained in this study was in agreement with that of cellulose extracted from pineapple core (Prakongpan et al., 2002). Detailed assessment of functional properties of the extracted cellulose will help in designing applications from the extracted polymer.

Table 2
Functional properties of extracted cellulose

Functional property	Extracted Cellulose	Commercial cellulose (Singanusong et al., 2014)
Bulked Density (g/mL)	0.40 ± 0.01	0.192 ± 0.01
Packed Density (g/mL)	0.81 ± 0.02	0.265 ± 0.07
Hydrated Density (g/mL)	2.42 ± 0.01	1.67 ± 0.36
Water Retention Capacity (WRC) (g/g)	2.62 ± 0.12	1.93 ± 0.82
Oil Retention Capacity (ORC) (g/g)	3.81 ± 0.12	3.17 ± 0.15
Emulsifying activity(EA)	59.11 ± 0.07	56.05 ± 0.06
Setting volume [(cm/cm) %]	15.3 ± 0.11	14.0 ± 0.16
*Data represents mean ± SD values performed in triplicate which is significant at P < 0.05.		

3.3. Characterization of extracted cellulose

3.3.1. FTIR spectrum of extracted cellulose

The FT-IR spectroscopy gives the information about the functional group and its property. The FTIR spectra of cellulose extracted from *water lettuce* sp. (Fig. 3) showed a broad peak at 3331.39 cm⁻¹, broad peak around 3500 – 3200 cm⁻¹ represents free O-H stretching of the OH group present in cellulose (Lu & Hsieh, 2010). The peak at 2853.03 cm⁻¹ in the spectrum represents the characteristic C-H stretch vibration of cellulose (Luzi et al., 2019; Mandal & Chakrabarty, 2011). The hemicellulose impurities from the extracted cellulose depict carbonyl aldehyde and ester (acetylated) groups, which are significantly determined by the C = O stretching vibration at 1744.95 cm⁻¹ (Reddy et al., 2009). The peak seen at 1609.29 cm⁻¹ corresponds to the absorption of water (Mandal & Chakrabarty, 2011). The occurrence of aromatic functional groups viz., methoxyl, and methylene groups in the side chains of lignin are referred to as the presence of C-H deformation and C = C ring stretching at 1457.93 cm⁻¹ in the spectra (Maheswari et al., 2012; Sain & Panthapulakkal, 2006). The peaks are not prominent as the extraction removed much of the lignin. The band at 1420.80 cm⁻¹ correlates to the CH₂ bending of cellulose (Jmel et al., 2019; Maheswari et al., 2012). Further, the presence of cellulose indicated by skeletal vibrations of C-C and C-O at 1316.56 cm⁻¹. In addition to these peak at 896.75 cm⁻¹ represent the β-glycosidic linkages between glucose molecules (Pappas et al., 2002; Sain & Panthapulakkal, 2006). FTIR confirms effective removal of hemicellulose and lignin contents by alkali treatment and bleaching of water lettuce.

3.3.2. SEM analysis of extracted cellulose

The morphological characterization of extracted cellulose using the SEM analysis shown in Fig. 4, where the fibers appeared glossy fiber-shaped texture with a smooth surface (Meng et al., 2019). The

morphological analysis confirms that the impurities are removed to an extent after bleaching. The surface morphology also indicates the presence of non-cellulosic impurities, which are mainly lignin, hemicellulose, pectin wax and other impurities (Chandrasekhar et al., 2004; Nurain et al., 2012), these are the natural binders of cellulose. It is also observed that the cellulose bundles are broken into small irregularly shaped fragments which happened during the delignification reactions.

3.3.3. XRD spectrum of extracted cellulose

The peak in the XRD data of extracted cellulose (Fig. 5), observed at 2θ (22.455°), is a characteristic peak observed in all cellulose XRD analysis (Elanthikkal et al., 2010; Wang et al., 2007). The high intensity peak at 11.3756° , 19.0336° and 33.6920° correlated to the structure of cellulose I, and are commonly found in plants (Bogolitsyn et al., 2014; Isogai et al., 1989; Visakh & Thomas, 2010; Zhang et al., 2020). The crystalline index was found to be 87.79%. The high crystalline

The effective removal of recalcitrant lignin, hemicellulose, and other non-cellulosic impurities by the mercerization and bleaching of water lettuce biomass aid in the generation of highly crystalline cellulose (Djalal & Klapötke, 2019; Meng et al., 2019).

3.3.4. TGA of extracted cellulose

As per TGA curve (Fig. 6), the initial weight loss by 8.820% occurred at 30°C . This reduction in weight can be attributed to loss of water content. Subsequent reduction by 60.53% was found to be highest, between the temperature range of $202.73\text{--}400.6^\circ\text{C}$. The greater reduction in weight occurred due to fast pyrolysis as a consequence of dehydration and degradation of cellulose. Further reduction by 10.36% at $400.6\text{--}789^\circ\text{C}$ occurred due to slow pyrolysis. The remaining residue of about 20.32% was due to the presence of lignin impurities (Kale et al., 2019; Yeng et al., 2015).

3.3.4. DSC spectrum of cellulose

When the cellulose sample extracted from water lettuce was subjected to high temperature (Fig. 7) the highest peak was obtained at 110.4°C which is lying in the glass transition range (100 to 180°C) of cellulose (Szcześniak et al., 2008). Other peaks are obtained at 65.4°C , 83.3°C and 157.9°C . The peak at 65.4°C may be due to the loss of water (Picker & Hoag, 2002). Other two peaks can be due to the pectin, hemicellulose and lignin impurities.

3.4. Biodegradation of cellulose sheets

Cellulose sheets were fabricated with incorporation of starch and dried (Fig. 8(a)). Further biodegradation study was carried out by packing the prepared CS in an inert matrix (Fig. 8(b) and burying it in garden soil followed by evaluation of degradation rate as a product of weight loss of the CS for a period of 4 weeks (28 days) by measuring weight loss every 7 days for further application and processing (Fig. 9).

Degradation of polymeric sheets by enzymes produced by soil microorganisms are basically a surface phenomenon as these enzymes cannot penetrate into the interiors of the polymeric sheets (Gouda et al., 2001). Thus weight loss can be successfully used as a measure to assess the enzymatic cleavage of

polymers. Microbial degradation of biopolymers usually proceeds through four phases. In the first phase fragmentation of polymers due to enzyme activity and effect of abiotic factors takes place and is combinedly called as bio deterioration phase. In the second depolymerization phase a gradual reduction in molecular weight of polymers occurs. The third phase of degradation involves assimilation of degradation products into the metabolism of microorganisms for structural and functional responsibilities. In the final mineralization stage of degradation complete oxidation of degradation products takes place (Lucas et al., 2008). The degradation (%) of CS drastically increased from week 1 (11.13 ± 0.83) to week 4 (78.22 ± 0.71). The degradation rate of CS was consistently higher than that of FP and CT sheets used for comparative study. Assessment of CT degradation rate of CT sheets were difficult as compared to FP and CS as the weight loss values were not showing much significant variation probably due to the inability of microbial enzymes to degrade the CT sheets due to their high molecular weight polypropylene composition. The high biodegradation rate of CS as compared to commercial FP as evident from (Fig. 9), may be attributed to the usage of starch as plasticizer as compared to glycerol in commercial FP. The results obtained from this study were promising to be extended as a packaging material due to their high degree of biodegradability. The results obtained in this study were in correlation with the biodegradation studies of cellulose films prepared from durian rind, where 100% biodegradation was observed within 4 weeks of soil burial (Zhao et al., 2019). In another study with nano cellulose films extracted from jackfruit peels, the cellulose films without any plasticizers incorporated reported high degree of biodegradation with 35 days of soil burial. Further their study reported that the moisture adsorption capacity of the plasticizers could be a key ingredient in determining the degradation rate of biopolymers (Reshmy et al., 2021). Similar pattern of biodegradation was reported with cellulose films prepared from banana pseudo stem when subjected to weight loss analysis after soil burial method for a period of 4 weeks (Ai et al., 2021). Cellulose biodegradation was reported to produce harmless end products like methane, CO₂ and water, the methane produced can be harnessed and integrated for bioenergy generation using modern technologies (Sharma et al., 2020).

4. Conclusion

The research interest on cost effective production of commercially important biopolymers called for the integration of waste valorization along with product development. The present work explores the possibility of extracting cellulose from a noxious aquatic weed, water lettuce that has a global distribution pattern resulting in numerous detrimental effects to aquatic ecosystems. In this method, although an eco-friendly method was attempted in a view to serve as an alternative to chemical extraction method, it was not found to be effective in cellulose recovery. Further screening for natural bleaching agents and optimization of the extraction process may improve the eco-friendly method for cellulose recovery. In this work, an attempt was made to make a biodegradable sheet with and without including plasticizers to improve its biodegradability without compromising the material properties pertaining to biodegradability. Soil burial method of the cellulose sheets clearly reflected the biodegradable nature of the developed sheets. Further studies on the mechanical properties and blending

with other natural polymers can improve the functional properties of the cellulose sheet and widens its application perspectives.

Declarations

Competing interests:

The authors declare no conflict of interest.

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Figures

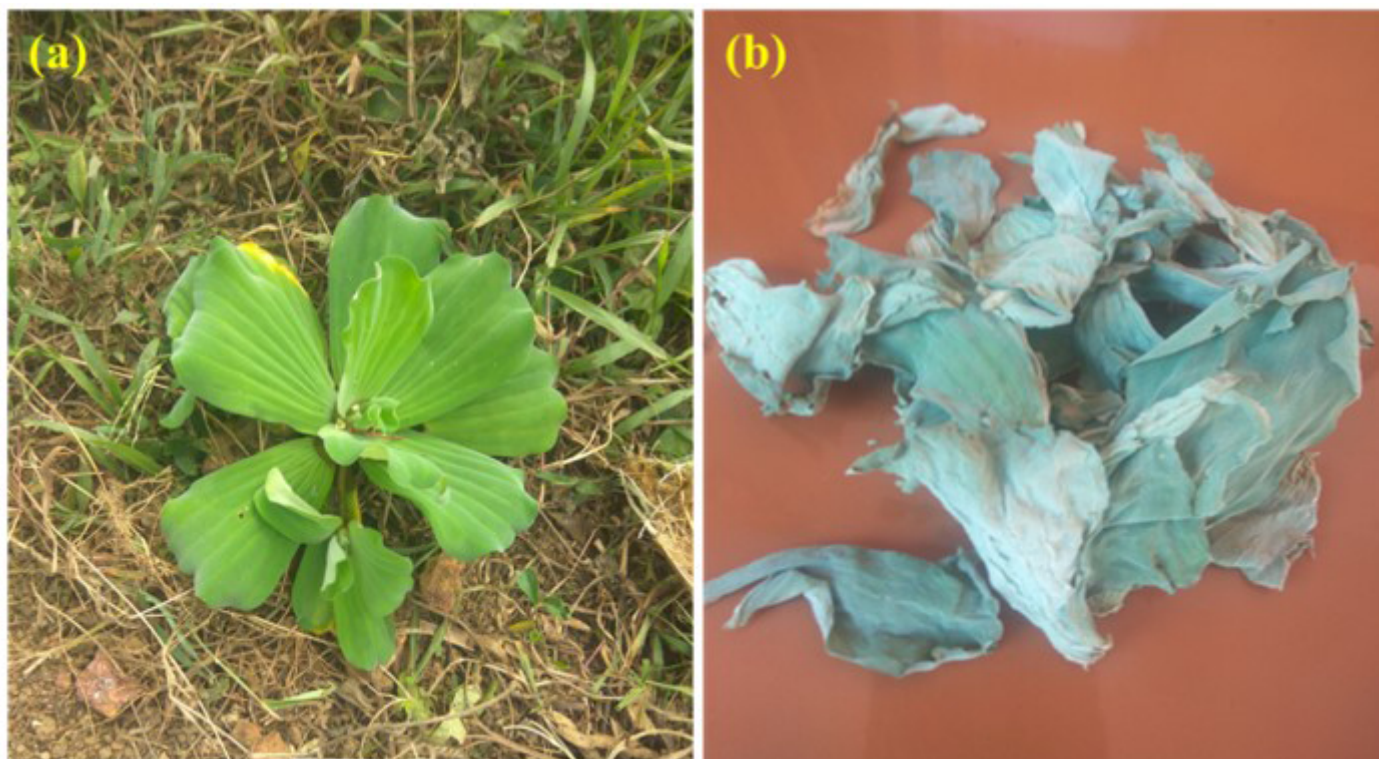


Figure 1

Collection of water lettuce (substrate) (a): Fresh water lettuce collected from the river (b) Water lettuce leaves after drying.

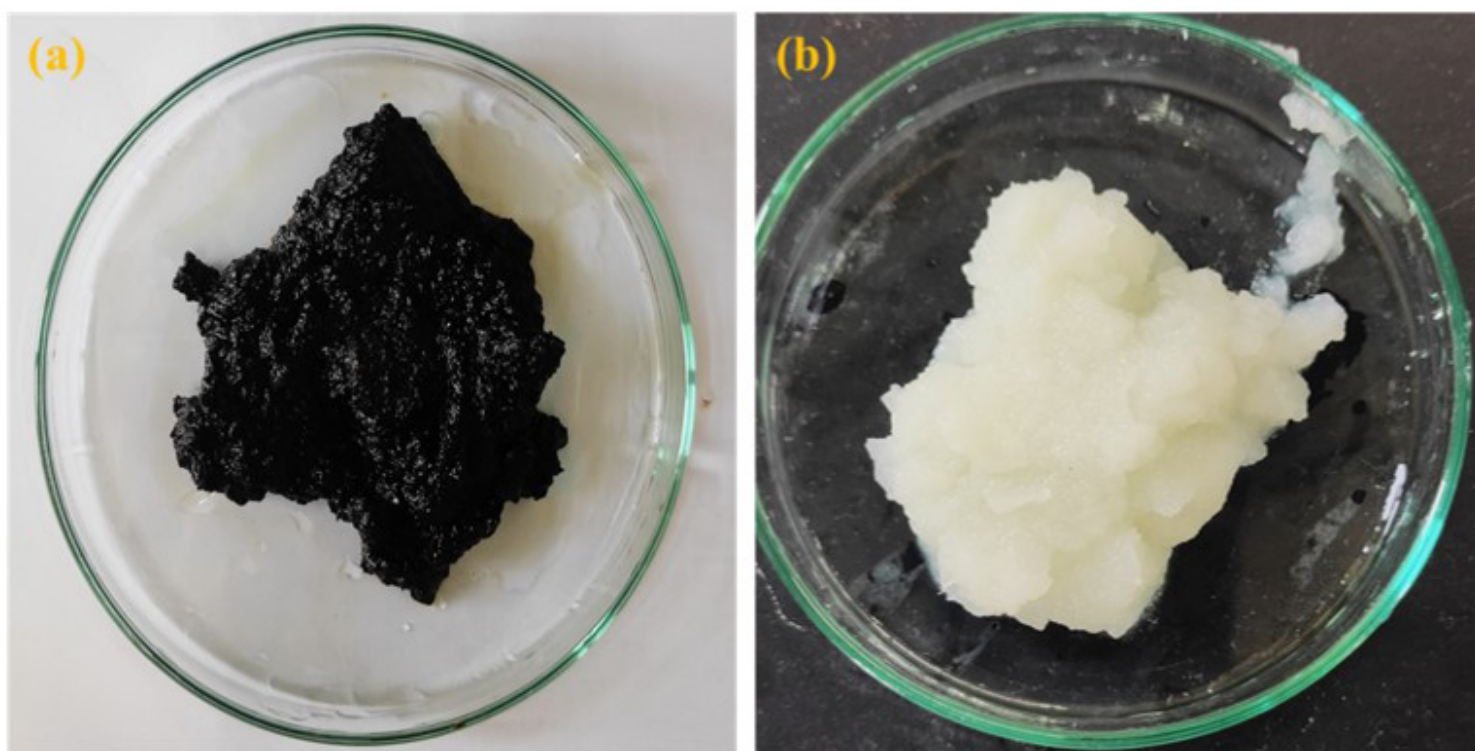


Figure 2

Extracted cellulose from water lettuce. (a): Water lettuce after alkali treatment. (b): Water lettuce sample after bleaching.

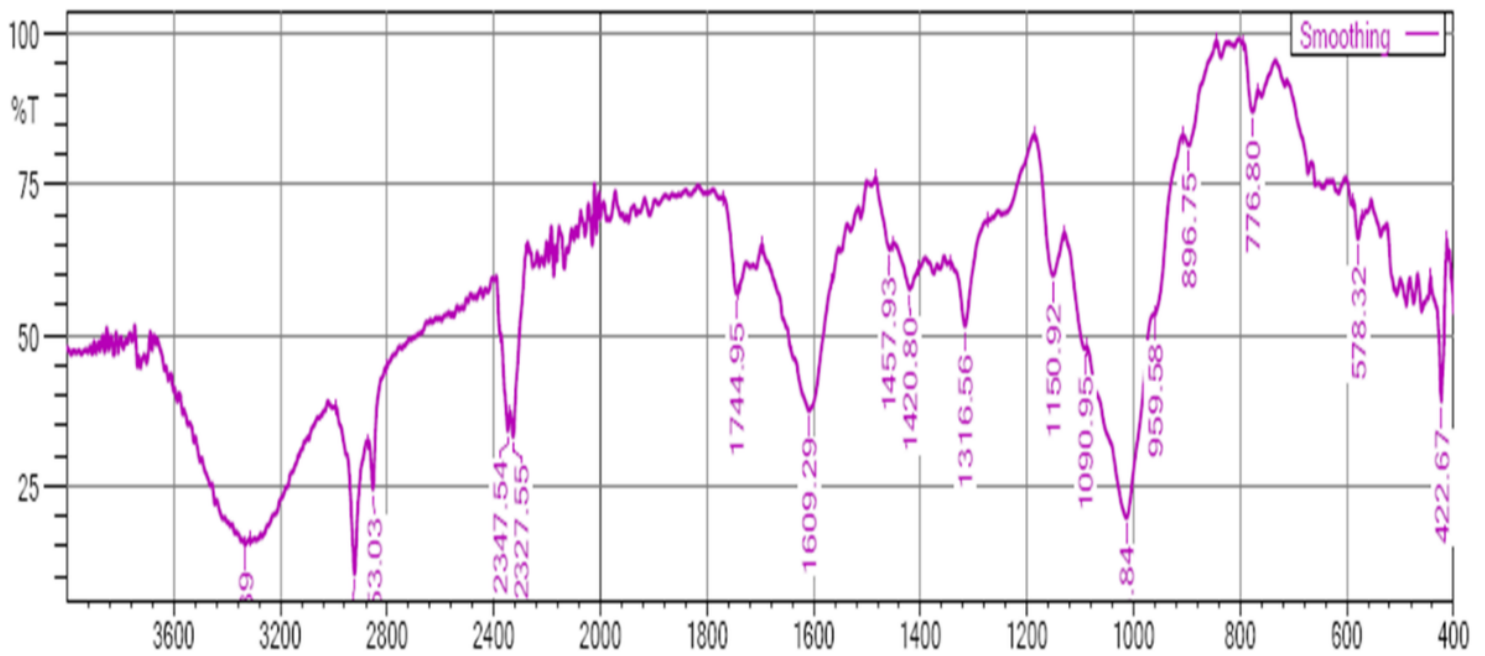


Figure 3

FT-IR Spectrum of extracted cellulose from water lettuce

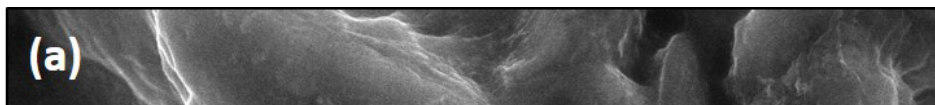


Figure 4

SEM image of extracted cellulose from water lettuce

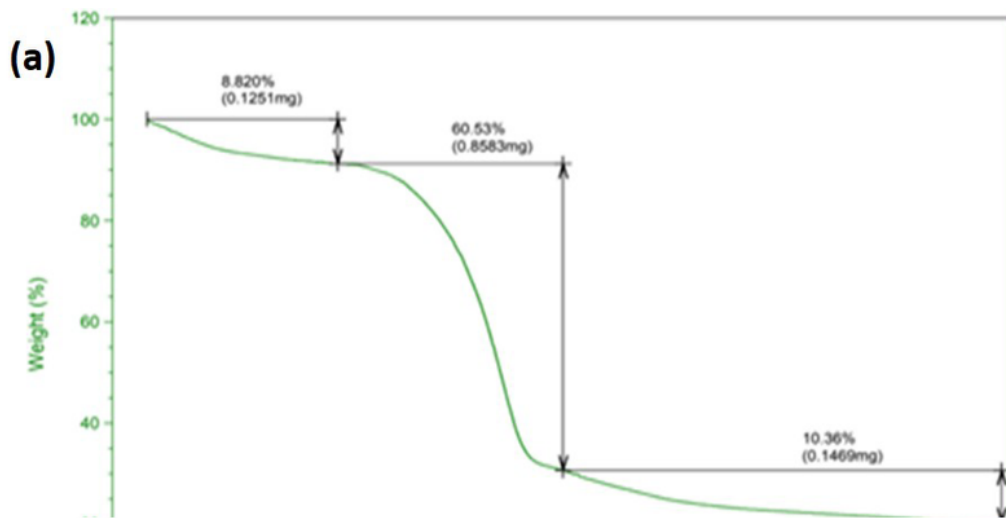


Figure 5

XRD spectrum of extracted cellulose from water lettuce



Figure 6

TGA spectrum of extracted cellulose from water lettuce

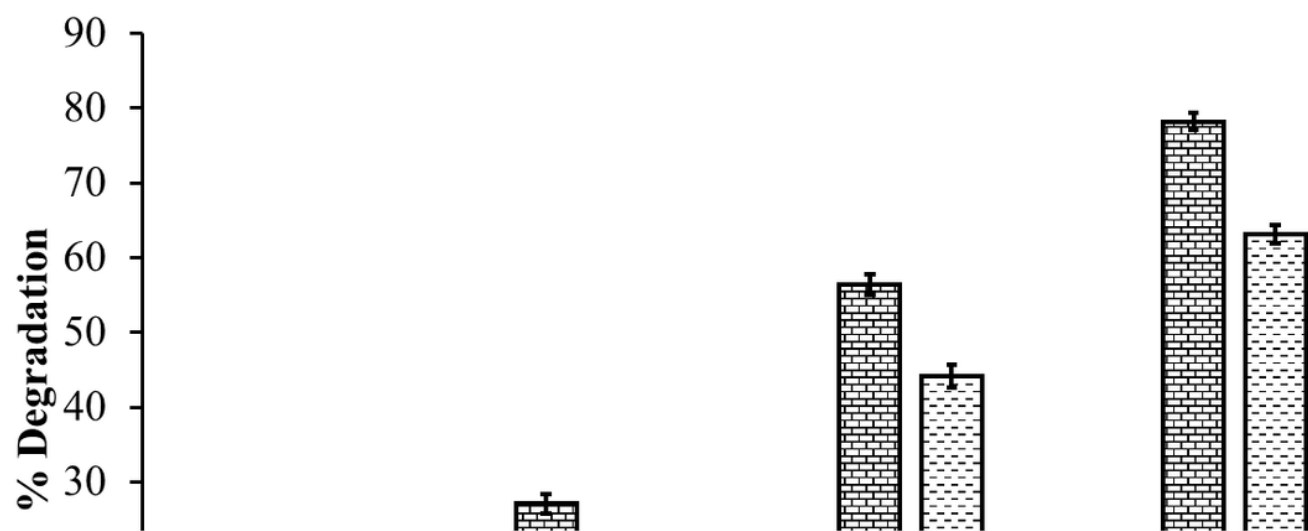


Figure 7

DSC spectrum of extracted cellulose from water lettuce

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Figure 8

Fabrication of cellulose biopolymer sheet from water lettuce (a): Fabricated cellulose sheet (b) Cellulose sheet packed in inert matrix

Image not available with this version

Figure 9

Degradation rate of the cellulose sheet synthesized from water lettuce. Cellulose from water lettuce (CS), acetate filter paper sheets (FP), and cellophane tapes (CT).