

Review Article

An In-Depth Review of Sustainable and Environmentally Friendly Pretreatment Techniques for Cellulose Extraction from Lignocellulosic Biomass and Their Uses

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Abstract

Lignocellulosic biomass (LCB), primarily composed of cellulose, hemicellulose, and lignin, represents a valuable renewable resource. Among these components, cellulose is the most abundant biodegradable polymer, with significant industrial potential. However, its efficient isolation remains a major challenge due to its strong association with hemicellulose and lignin within the complex LCB structure. Effective pretreatment strategies are essential to overcome these structural barriers and facilitate cellulose extraction. Conventional pretreatment methods, while effective, often rely on harsh chemicals, elevated temperatures, and high energy inputs, leading to increased costs, low yields, and the formation of fermentation inhibitors, which hinder sustainable biomass utilization. In response to these limitations, environmentally friendly pretreatment approaches have been developed to enhance cellulose separation while minimizing ecological and economic drawbacks. These green methods prioritize efficient delignification, reduced solvent consumption, lower environmental impact, and alignment with sustainable technological advancements. This review explores the application of eco-friendly pretreatment techniques for cellulose isolation from diverse lignocellulosic biomass sources. It further highlights the role of cellulose as a crucial feedstock in the bioeconomy, emphasizing its versatility across various industrial sectors. By advancing sustainable cellulose extraction methods, this research contributes to the broader goal of developing environmentally responsible and economically viable biorefinery processes.

Keywords

Bio-based, Lignocellulosic Biomass, Cellulose, Green Pretreatment, Conventional Pretreatment, Cellulose Derivatives, Environmental Sustainability, Delignification

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1. Introduction

The increasing demand for bio-based, carbon-neutral, and renewable feedstocks to replace fossil fuels across various industries has intensified research interest in sustainable feedstock sources [1]. Lignocellulosic biomass, an abundant and renewable plant-based resource, holds significant potential in this context. As illustrated in Figure 1, its complex structure primarily consists of the polysaccharide's cellulose and hemicellulose, along with the phenolic macromolecule lignin, in addition to minor constituents such as proteins, lipids, and inorganic compounds [2-4].

The composition of lignocellulosic biomass varies, with cellulose accounting for approximately 35–55% by mass, hemicellulose 20–40%, and lignin 10–25%, alongside other polar and non-polar compounds [5]. Furthermore, its elemental composition can be categorized into major elements (e.g., C, H, O, N, K, and Ca), minor elements (e.g., Mg, Al, Si, P, Cl, Na, S, and Fe), and trace elements (e.g., Mn and Ti). Understanding the structural and chemical complexity of lignocellulosic biomass is crucial for optimizing its utilization in sustainable biorefinery processes, advancing the transition toward a circular bioeconomy [6, 7].

Cellulose, the primary structural component of lignocellulosic biomass, is a homopolysaccharide composed of D-glucose monomer chains linked by β -1,4 glycosidic bonds. These chains are stabilized through hydrogen bonding and van der

Waals forces [2], contributing to cellulose's highly ordered and crystalline nature. The fundamental repeating unit of cellulose is cellobiose (D-glucopyranosyl- β -1,4-D-glucopyranose), and its interaction with lignin is mediated by hemicellulose through hydrogen and covalent bonding [8].

Hemicellulose, in contrast, is a branched heteropolysaccharide composed of a diverse range of monosaccharides, including five-carbon sugars (xylose and arabinose) and six-carbon sugars (mannose, glucose, and galactose), along with carboxylic acids such as mannuronic and galacturonic acids. Beyond its structural role, hemicellulose also facilitates the deposition of lignin monomers within the secondary cell wall [9].

Lignin, a complex and amorphous polymer, consists of monolignol precursors such as p-coumaryl alcohol, sinapyl alcohol, and coniferyl alcohol [9, 10]. It forms an extensively crosslinked macromolecular network through the polymerization of phenylpropanoid monomers. As a hydrophobic and highly recalcitrant biopolymer, lignin serves as a protective barrier, limiting cellulose accessibility by anchoring hemicellulose to the cellulose framework within the cell wall [10, 11]. Its resistance to hydrolysis presents a significant challenge in biomass processing, necessitating efficient pre-treatment strategies to enhance cellulose availability for industrial applications [10-14].

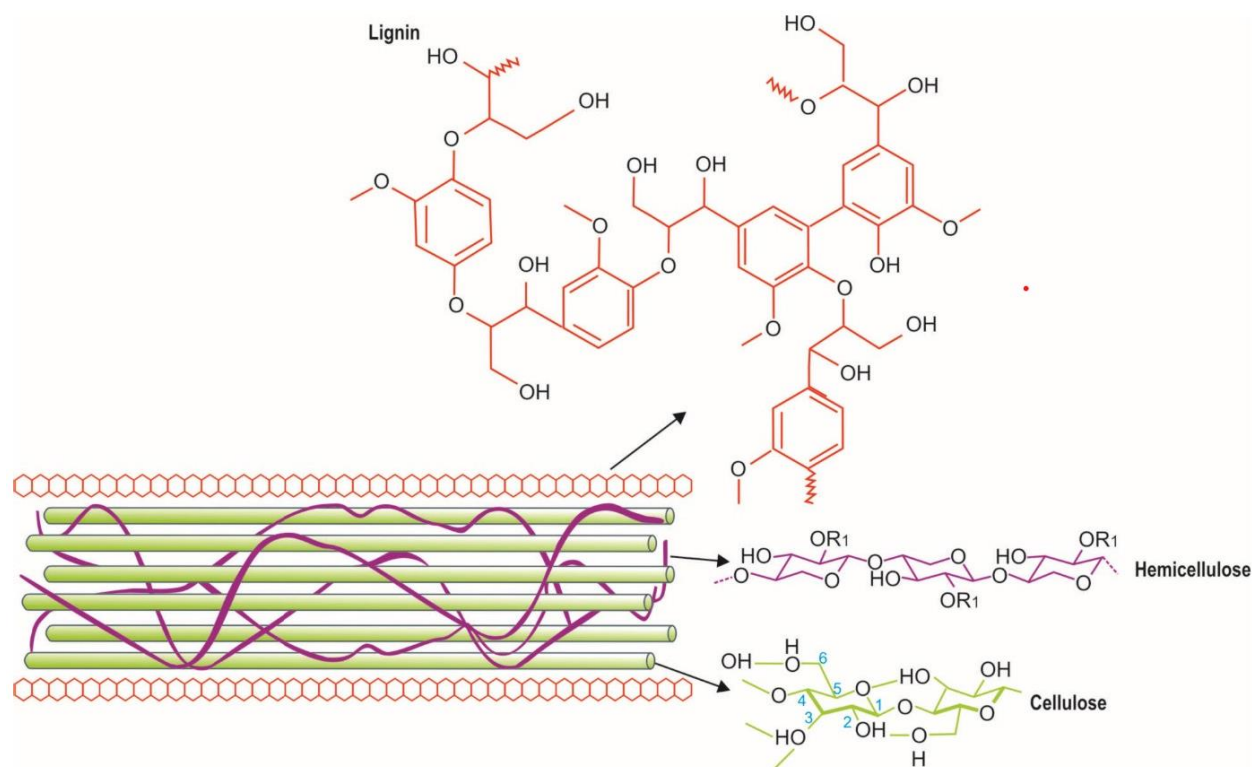


Figure 1. Lignocellulose composition and organization [3].

Cellulose, a polysaccharide composed of β -D-glucopyranose units linked by β -(1,4) glycosidic bonds, has garnered considerable interest as a sustainable feedstock for biofuels, chemicals, and bioproducts due to its biodegradability, renewability, and exceptional mechanical properties [12]. However, its extraction from lignocellulosic biomass remains challenging, as cellulose is tightly embedded within a matrix of lignin and hemicellulose, limiting its direct enzymatic hydrolysis into glucose [15].

To enhance cellulose accessibility, a pretreatment process is necessary to disrupt these structural barriers, facilitate lignin dissolution, and reduce cellulose crystallinity, thereby improving enzymatic hydrolysis efficiency [16]. While conventional pretreatment techniques often involve harsh conditions, the development of environmentally friendly alternatives has become a priority to mitigate their ecological impact. Green pretreatment strategies, including organosolv pretreatment, biological methods, supercritical fluid applications, and deep eutectic solvents, offer sustainable approaches for cellulose isolation. Additionally, process intensification techniques such as microwave-assisted, ultrasonication, and cold plasma-assisted pretreatment have emerged as innovative solutions to enhance efficiency while minimizing environmental and economic burdens [17, 18].

1.1. Structure and Properties of Cellulose

Cellulose is a polysaccharide that serves as a primary structural component of plant cell walls, consisting of β -D-glucose units connected by β -1,4-glycosidic bonds. This configuration results in the formation of linear, semicrystalline fibrillar structures [19-21]. Cellulose is further organized into ultrafine fibril bundles, which exhibit alternating crystalline and amorphous regions, typically ranging in size from 2 to 20 nm [22, 23].

The highly ordered crystalline regions are characterized by tightly packed cellulose chains, reinforced by extensive intermolecular and intramolecular hydrogen bonding [24]. This structural organization imparts significant mechanical strength to cellulose while rendering it insoluble in water and most organic solvents. However, its close association with a surrounding hemicellulose-lignin matrix presents challenges for its fractionation and subsequent valorization in industrial

applications [25].

Despite these structural barriers, certain microorganisms and enzymatic systems have evolved the capability to hydrolyze cellulose into glucose, a critical process for biofuel production, bioproduct synthesis, and other industrial applications. Understanding the physicochemical properties of cellulose is essential for optimizing its utilization in sustainable biorefinery processes [26].

1.2. Sources of Cellulose

Cellulose is abundantly available from various lignocellulosic biomass sources, including wood and non-wood plants, agricultural residues, and municipal or industrial lignocellulosic waste [27]. The cellulose, hemicellulose, and lignin composition of these biomass sources varies depending on their origin [27], as summarized in Table 1.

Wood-derived cellulose can be obtained from both hardwood species (e.g., oak, maple, and birch) and softwood species (e.g., pine, spruce, and fir). Non-wood plant sources include fiber-rich crops such as cotton, hemp, flax, jute, and kenaf, which are cultivated primarily for their high cellulose content. Additionally, agricultural residues such as coconut coir, wheat straw, maize stover, rice straw, and empty fruit bunches from palm trees serve as valuable sources of cellulose [27].

Beyond plant-derived sources, cellulose can also be recovered from municipal and industrial waste streams, including food and beverage industry byproducts, spent grains from breweries, sugarcane bagasse, and papermaking sludge. Furthermore, certain microorganisms, including specific strains of bacteria, algae, and fungi, are capable of synthesizing cellulose, offering alternative and sustainable production pathways. Understanding these diverse sources is essential for optimizing cellulose extraction and utilization in various industrial applications [28].

The composition of lignocellulosic biomass varies significantly depending on the source, influencing its suitability for cellulose extraction and industrial applications. Table 1 presents the cellulose, hemicellulose, and lignin content of different biomass sources, highlighting their structural diversity.

Table 1. Composition of Lignocellulosic Biomass from Various Sources.

Source	Cellulose (%)	Hemicellulose (%)	Lignin (%)	References
Brewer spent grain	23.1	22.9	19.0	[29]
Corn stover	31.5	18.0	14.1	[30]
Poplar sawdust	46.2	19.3	26.15	[31]
Sugarcane bagasse	44	28	21	[32]
Wheat straw	29.9	13.8	32.8	[30]

Source	Cellulose (%)	Hemicellulose (%)	Lignin (%)	References
Eucalyptus globulus wood	14.1	20.5	50	[33]
Raw kenaf	21.2	22.8	45.8	[34]
Jute	12.24	31.58	42.5	[35]
Beechwood chips	22.3	25.9	41.6	[27]

These variations in composition underscore the importance of selecting appropriate biomass sources for cellulose extraction. For instance, poplar sawdust and sugarcane bagasse contain relatively high cellulose content, making them promising candidates for biorefinery applications. In contrast, biomass sources with higher lignin content, such as *Eucalyptus globulus* wood and jute, require more intensive pretreatment strategies to enhance cellulose accessibility. Understanding these compositional differences is crucial for optimizing biomass utilization in sustainable industrial processes.

2. Methods of Cellulose Extraction from Lignocellulosic Biomass

The extraction of cellulose from plant cell walls requires the separation of cellulose from lignin and hemicellulose through various physical, chemical, physicochemical, and biological pretreatment methods [36]. The efficiency and quality of cellulose extraction depend on several factors, including the biomass source, pretreatment strategy, solvent selection, process conditions (temperature, pressure, and time), and mechanical agitation [37].

The extraction process generally involves three key stages: biomass conditioning, pretreatment, and purification or bleaching to eliminate residual lignin and hemicellulose [38]. Biomass conditioning includes washing, size reduction, and refluxing with ethanol and deionized water to remove impurities, which enhances surface area and facilitates the breakdown of lignin barriers, improving accessibility for subsequent treatments [38].

Pretreatment techniques play a crucial role in disrupting the lignocellulosic matrix and solubilizing lignin and hemicellulose while preserving cellulose integrity [39-51]. These methods include acid or alkaline pulping, oxidative treatments using ozone or peroxide, steam explosion, organosolv pretreatment, and ultrasound-assisted processes. Following pretreatment, bleaching agents such as hypochlorite or peroxide are applied to oxidize and remove residual lignin and other degraded compounds. This bleaching process is often repeated to obtain high-purity, white cellulose fibers [52].

Once purified, the extracted cellulose is dried and processed into bio-based products, contributing to the development of sustainable materials and bioeconomy applications.

Optimizing these extraction techniques is essential for enhancing cellulose yield, minimizing environmental impact, and ensuring efficient utilization of lignocellulosic biomass.

2.1. Alkaline Pretreatment

Alkaline pretreatment is a widely used and cost-effective method for processing lignocellulosic biomass, employing alkaline reagents to disrupt the lignin-hemicellulose complex and enhance cellulose accessibility for enzymatic hydrolysis [53]. This process primarily involves the cleavage of intermolecular ester bonds between lignin and hemicellulose, leading to their solubilization and improved cellulose exposure [54]. The effectiveness of alkaline pretreatment is influenced by key parameters such as the type and concentration of the alkaline reagent, reaction time, and processing temperature [53].

Among various alkaline reagents, sodium hydroxide (NaOH) is the most commonly utilized due to its high efficiency, low cost, and ease of handling [55-63]. Alkaline pretreatment offers several advantages, including effective lignin removal, which enhances enzymatic hydrolysis by improving enzyme accessibility and reducing enzyme loading requirements, thereby lowering overall processing costs. Additionally, the partial hydrolysis of hemicellulose during this process yields fermentable sugars that can be utilized for biofuel production [40].

Despite its benefits, alkaline pretreatment presents certain challenges. The process can be non-selective, leading to the partial degradation and loss of cellulose and hemicellulose. The use of strong alkalis, such as NaOH and ammonium hydroxide, raises environmental concerns related to chemical handling, storage, and waste disposal. Moreover, harsh alkaline conditions can negatively impact cellulase enzymes, leading to enzyme deactivation and reduced hydrolysis efficiency. Additionally, the generation of inhibitory compounds, including phenolics, furans, and organic acids, may hinder enzymatic hydrolysis and lower overall biomass conversion efficiency. Addressing these limitations through process optimization and environmentally sustainable alternatives is crucial for improving the viability of alkaline pretreatment in biorefinery applications.

2.2. Acid Pretreatment

Acid pretreatment is a widely adopted technique for cellulose isolation from lignocellulosic biomass, particularly in biofuel production. This method involves the application of acids to disrupt the lignocellulosic structure by solubilizing hemicellulose and lignin, thereby improving cellulose accessibility for enzymatic hydrolysis into fermentable sugars. Both mineral acids (such as hydrochloric, phosphoric, and sulfuric acids) and organic acids (including acetic, citric, and formic acids) are commonly employed in this process [64-70].

Dilute mineral acids are preferred over concentrated acids due to their ability to efficiently hydrolyze glycosidic bonds, facilitating hemicellulose solubilization while minimizing sugar degradation. The process is typically conducted at temperatures ranging from 160–220 °C, with acid concentrations between 0.05–5%, and relatively short reaction times [71]. Among various acids, dilute sulfuric acid has demonstrated high efficacy in pretreating a range of biomass types, such as distillery stillage, bamboo, and corn stover, due to its strong lignin and hemicellulose solubilization capacity [72-76]. For instance, Sahoo et al. [76] reported that pretreating wild rice grass with 2% sulfuric acid at a 10% biomass loading resulted in optimal sugar release, which was subsequently utilized for enzymatic hydrolysis and bioethanol production.

Acid pretreatment offers several advantages, including effective lignin removal, enhanced cellulose accessibility, and increased biomass digestibility. Additionally, it modifies the crystalline structure of cellulose and partially hydrolyzes hemicellulose, improving enzyme activity and sugar conversion efficiency, which is essential for biofuel synthesis.

Despite these benefits, acid pretreatment presents certain limitations. A neutralization step is required to mitigate acid residues, generating salts and degrading sugars, leading to the formation of fermentation inhibitors such as furfural. The corrosive nature of acids necessitates the use of specialized equipment, increasing maintenance costs and infrastructure investments. Furthermore, acid recovery is economically challenging, and the environmental impact of acid waste disposal highlights the need for more sustainable and efficient pretreatment alternatives. Developing eco-friendly approaches with minimal byproduct formation is crucial for improving the feasibility of acid pretreatment in biorefinery applications [16, 77].

3. Green Methods for Cellulose Extraction from Lignocellulosic Biomass

Green methods for cellulose extraction aim to utilize environmentally friendly solvents and processing conditions to minimize hazardous waste generation and reduce greenhouse gas emissions. The core objective of these sustainable ligno-

cellulose biomass fractionation techniques is to mitigate pollution and the production of toxic byproducts during the extraction of biomass components, while also developing cost-effective and eco-friendly approaches. These methods focus on enhancing cellulose yield while minimizing the generation of harmful waste and promoting resource efficiency [58].

A process, solvent, or product is considered "green" if it minimizes its environmental impact throughout its entire lifecycle, from production to disposal. The adoption of green methods offers several benefits, including a reduced environmental footprint, lower energy consumption, and limited reliance on harmful chemicals [73].

Several innovative green approaches are being explored in lignocellulose biomass pretreatment. These include the use of organosolvents, deep eutectic solvents, and supercritical fluids, as well as non-chemical techniques such as cold plasma, enzymatic treatments, and physical treatments like ultrasound-assisted and microwave-assisted pretreatment. These methods not only offer more sustainable alternatives to traditional pretreatment processes but also show promise in improving the efficiency and effectiveness of cellulose extraction from lignocellulosic biomass, making them valuable components of a sustainable bioeconomy [58].

3.1. Organosolvent Pretreatment

Organosolvent pretreatment is an emerging green technique for fractionating LCB, which involves the use of organic solvents, often in combination with water, to dissolve and remove lignin while retaining high-purity cellulose. This process typically occurs at temperatures between 150 °C and 200 °C, with or without the addition of acid catalysts, such as sulfuric or acetic acid, to optimize lignin removal and preserve the integrity of cellulose [78, 79]. Commonly used solvents include ethanol, methanol, acetone, and ethylene glycol, with the addition of acid catalysts enhancing the process, although careful control is required to avoid excessive cellulose degradation [78].

Organosolvent pretreatment offers several advantages, including efficient separation of lignocellulosic biomass into its constituent components—cellulose, lignin, and hemicellulose. It also provides the benefit of solvent recovery and reuse, which enhances the sustainability of the process. Additionally, this method improves cellulose accessibility for subsequent enzymatic saccharification, leading to higher yields of fermentable sugars, making it a valuable approach for biofuel and bioproduct production [80, 81]. Organosolv pretreatment is also versatile, as it can be applied to various biomass feedstocks, reduces the need for excessive enzyme and chemical inputs, and helps lower energy consumption while minimizing the formation of fermentation inhibitors [82].

Despite these advantages, the organosolvent pretreatment process faces some challenges. The high cost of solvents and the energy-intensive nature of solvent recovery are significant

economic hurdles. To improve the cost-effectiveness of this method, effective solvent recycling strategies are essential. Additionally, care must be taken to ensure complete removal of solvents from the system, as residual solvents can interfere with downstream processes such as microbial growth, enzymatic hydrolysis, and fermentation, potentially reducing overall process efficiency. Further research into optimizing solvent recovery and enhancing process efficiency is needed to make organosolvent pretreatment a more economically viable and sustainable solution for biomass processing [81, 82].

3.2. Steam Explosion

Steam explosion (SE) is a thermomechanical pretreatment method that utilizes high-pressure steam, shearing forces, and organic acids to break down lignocellulosic biomass. This process involves exposing the biomass to high-pressure steam, followed by rapid decompression to atmospheric pressure, which disrupts hemicellulose-lignin linkages and enhances the accessibility of crystalline cellulose for enzymatic and acid hydrolysis [83-85]. The SE process consists of two main phases: explosive decompression and vapocacking, which lead to structural modifications in the biomass. These modifications include the hydrolysis of hemicellulose, changes in cellulose crystallinity, and alterations in the chemical structure of lignin, ultimately improving the efficiency of enzymatic hydrolysis and increasing fermentable sugar yields [86, 87].

The advantages of steam explosion include efficient solubilization of lignin, removal of impurities such as ash and salts, and enhanced cellulose accessibility for enzymatic saccharification. These benefits make steam explosion a promising method for pretreating lignocellulosic biomass, especially for biofuel production [87, 88].

However, steam explosion also presents some limitations. The process only partially disrupts lignin-carbohydrate linkages, which may not fully separate lignin from cellulose and hemicellulose. Moreover, the treatment can generate fermentation inhibitors, such as acetic acid, furfural, and hydroxymethylfurfural (HMF), which can negatively affect subsequent fermentation and hydrolysis steps. Additionally, steam explosion is a nonselective method, impacting all biomass components. This nonselectivity complicates the efficient fractionation of lignin, cellulose, and hemicellulose, potentially reducing cellulose yield and quality. Further optimization of the steam explosion process is needed to minimize these drawbacks and improve its overall efficiency in biomass pretreatment [84, 85].

3.3. Cold Plasma

Plasma, often referred to as the fourth state of matter, is a partially ionized gas consisting of ions, electrons, radicals, and neutral particles. This reactive environment is beneficial

for modifying the surface properties of plant fibers. Plasma can be categorized into two types: thermal plasma, which operates at high temperatures up to 4000 °C, and nonthermal (or cold) plasma (CP), which operates at ambient temperatures (25–100 °C) and is more suitable for low-temperature processing. Cold plasma has found applications in diverse fields, including microbial deactivation, concentration of bioactive substances, and enhancing antioxidant properties in food [89-91].

In the context of lignocellulosic biomass pretreatment, cold plasma offers an environmentally friendly and non-toxic approach to addressing the recalcitrance of biomass [92, 93]. Cold plasma treatment has been shown to improve various biomass processing stages, such as delignification, hydrolysis of cellulose into fermentable sugars, and biomass liquefaction, all of which are advantageous for the production of renewable fuels and chemicals. By creating a reactive environment, cold plasma enhances the accessibility and reactivity of biomass components, facilitating more efficient downstream processing [94-102].

Despite these advantages, there are challenges associated with cold plasma treatment. One significant drawback is the high energy consumption required to generate and maintain the plasma, which can increase operational costs [103]. Additionally, the complex equipment needed for cold plasma generation adds to the process's overall cost. Furthermore, while cold plasma can effectively modify biomass, the generation of reactive species—such as free radicals, ions, and UV radiation—during treatment can lead to the unintended degradation of biomass components, potentially causing the breakdown of lignocellulosic materials [92]. Thus, optimizing cold plasma parameters and balancing energy efficiency with biomass preservation remains an area of ongoing research.

3.4. High-Pressure Processing

High-pressure processing (HPP), also referred to as high hydrostatic pressure (HHP) or ultrahigh-pressure processing (UHP), is a nonthermal technology that operates at pressures ranging from 100 to 800 MPa, with capabilities extending up to 6000 atmospheres [104, 105]. This method is based on two fundamental principles: the Le Chatelier principle, which posits that pressure induces structural changes that reduce volume, and the isostatic principle, ensuring uniform pressure distribution across the material, regardless of its shape or size.

HPP has gained attention as an effective pretreatment for lignocellulosic biomass, particularly for enhancing its susceptibility to enzymatic hydrolysis, thus improving the conversion of biomass into fermentable sugars [106]. This method is particularly effective in disrupting lignin structures in highly recalcitrant biomass, such as wood, making it more amenable to subsequent enzymatic processes.

However, the application of HPP presents several challenges. One significant limitation is the high energy consumption required to generate and maintain such high pres-

tures. Additionally, there is a risk of biomass degradation during the process, which may affect the quality and yield of the final product. Scalability remains another challenge, as the equipment required for HPP is expensive and may not be easily adaptable for large-scale biomass processing. These factors contribute to the high operational costs of the method, limiting its widespread adoption for industrial-scale biomass processing. Further research and optimization are needed to address these challenges and improve the cost-effectiveness of HPP for large-scale applications [105].

3.5. Ultrasound-Assisted Pretreatment

Ultrasound-assisted pretreatment involves the application of high-frequency sound waves that propagate compression and rarefaction through a medium, generating acoustic cavitation. This phenomenon leads to localized high temperatures and pressures, producing reactive species such as free radicals ($\text{OH}^\cdot$, $\text{H}^\cdot$, H_2O_2) that enhance chemical reactions [107, 108]. Ultrasound treatment disrupts the cell walls of plant materials, reduces particle size, and degrades the plant matrix, thus improving mass transfer and increasing the efficiency of lignin removal when combined with solvents [109-112].

Research on ultrasound-assisted pretreatment has highlighted its effectiveness in lignin solubilization, demonstrating that ultrasound can significantly reduce the time and temperature required for lignin removal compared to conventional methods [113, 114]. Additionally, this technique has been shown to enhance the thermal stability of cellulose, with higher decomposition temperatures observed in cellulose extracted through ultrasound treatment [115].

However, there are challenges associated with ultrasound-assisted pretreatment. One primary limitation is the high energy consumption required to generate ultrasound waves, which can increase operational costs. Furthermore, scaling the process for industrial applications requires significant engineering modifications and process optimization to ensure efficiency and cost-effectiveness at larger scales. Despite these challenges, ongoing research into ultrasound-assisted pretreatment holds promise for improving the efficiency of lignocellulosic biomass fractionation for biofuel and bioproduct production [113].

3.6. Microwave-Assisted Pretreatment

Microwave-assisted pretreatment employs microwave energy to heat solvents and plant tissues, facilitating the breakdown of lignocellulosic biomass. The process is driven by ionic conduction and dipole rotation, where microwave energy interacts with polar molecules in the biomass, resulting in rapid heating and structural degradation [116, 117]. This technique enhances cellulose accessibility for enzymatic or chemical hydrolysis by partially breaking down lignin and disrupting the biomass structure. The effectiveness of the process is influenced by factors such as biomass type, mois-

ture content, particle size, solvent type, microwave power, and exposure time.

Research has demonstrated that microwave-assisted pretreatment is effective for a variety of biomass types, including softwoods, *Theobroma cacao* husk, and oil palm waste [116, 118]. Compared to traditional pretreatment methods, microwave-assisted pretreatment offers several advantages: it enables rapid and uniform heating, reduces pretreatment time, lowers energy consumption, and minimizes the need for chemicals, thereby reducing the environmental footprint [119]. Additionally, this method allows for selective targeting of specific biomass components, optimizing biofuel and biochemical yields, and supports real-time process monitoring for continuous process optimization.

Despite these advantages, several challenges remain. Uneven heating and limited penetration depth of microwave energy can lead to inconsistent treatment, affecting the quality of the pretreated material. Safety concerns related to microwave equipment and potential issues with scalability further complicate its widespread application. Additionally, the high costs associated with microwave equipment and energy consumption, coupled with the potential degradation of biomass components due to microwave-induced heating, can negatively impact the overall efficiency of the pretreatment process. Further research and optimization are required to address these limitations and enhance the viability of microwave-assisted pretreatment for large-scale applications [120].

3.7. Deep Eutectic Solvents (DESs)

Deep eutectic solvents (DESs) represent a promising class of environmentally friendly alternatives to ionic liquids for the pretreatment and delignification of LCB. DESs are typically formed by combining a hydrogen bond donor (HBD) and a hydrogen bond acceptor (HBA) in specific molar ratios, followed by heating to create a eutectic mixture at temperatures ranging from 70 °C to 100 °C [121]. This interaction leads to the disruption of the biomass structure through hydrogen bonding, weakening intermolecular forces within cellulose, hemicellulose, and lignin. As a result, DESs are effective in dissolving and converting lignocellulosic components [121].

DESs offer several advantages as a green solvent, including low toxicity, biodegradability, and ease of synthesis. Their ability to enhance biomass conversion rates and efficiently dissolve a variety of biomass components makes them a promising alternative for biomass pretreatment [122]. DESs have demonstrated efficacy in processes such as biomass delignification, cellulose extraction, and the purification of biopolymers from materials like wheat straw, softwood, and oil palm empty fruit bunches [118, 123, 124].

Compared to traditional pretreatment methods, DESs help reduce energy consumption by operating at lower temperatures and pressures, minimizing biomass degradation, and producing fewer inhibitory compounds, which enhances

fermentation efficiency. Furthermore, DESs are capable of preserving the crystalline structure of cellulose and can be recycled, leading to lower operational costs [121]. Despite these advantages, the use of DESs presents certain challenges, such as the complexity involved in solvent design, high synthesis costs, limited biomass penetration, and difficulties in scaling up and optimizing the process. Additionally, issues related to the handling, recovery, and disposal of DESs remain important considerations [121]. Further research is needed to address these limitations and improve the feasibility of DESs for large-scale biomass processing.

3.8. Supercritical Fluid Pretreatment

Supercritical fluid extraction (SFE) leverages the unique characteristics of supercritical fluids—substances that are above their critical temperature and pressure—to extract components from lignocellulosic biomass. Among supercritical fluids, carbon dioxide (CO₂) is the most commonly employed due to its relatively low critical temperature (31.1 °C) and pressure (73.8 bar), which make it a safe and accessible choice for biomass pretreatment [125, 126]. In its supercritical state, CO₂ exhibits both gas-like and liquid-like properties, allowing it to effectively penetrate porous biomass structures, making it an ideal solvent for SFE [127]. Other fluids such as ethanol, water, and phenol can also be utilized, depending on the specific application.

The primary advantages of SFE include the selective degradation of lignin while preserving cellulose and hemicellulose, which enhances the efficiency of subsequent conversion processes. This method significantly reduces the need for chemical additives, minimizes the formation of inhibitory byproducts, and improves the reactivity of biomass, which ultimately enhances product quality. Additionally, SFE offers precise control over extraction conditions by adjusting temperature and pressure, and it can reduce enzyme requirements, improving overall process efficiency [126].

However, despite these advantages, the main limitations of SFE include the high capital and operational costs associated with the specialized equipment and the complexity of the process [126]. These factors may limit its widespread adoption for large-scale biomass processing, although ongoing research into cost-reduction strategies and optimization techniques may improve its commercial feasibility.

3.9. Biological Pretreatment

Biological pretreatment, also known as biodelignification, utilizes microorganisms or enzymes to degrade lignin in lignocellulosic biomass, providing an environmentally sustain-

able alternative to conventional chemical methods. Among microorganisms, white-rot fungi (WRF) are particularly effective as they can completely degrade both lignin and polysaccharides. In contrast, brown-rot and soft-rot fungi primarily degrade polysaccharides with minimal impact on lignin. Certain bacterial strains, such as *Clostridium* sp., *Cellulomonas* sp., and *Streptomyces* sp., have also demonstrated lignocellulose-degrading capabilities [128-130].

The success of biological pretreatment depends on several factors, including the selection of appropriate microorganisms, optimization of environmental conditions (such as moisture, temperature, and aeration), and substrate characteristics (such as particle size, pH, and nutrient availability). Solid-state fermentation (SSF) is commonly employed to facilitate biological delignification [128], while enzymatic pretreatment involves directly applying ligninolytic enzymes like laccase and lignin peroxidase to selectively degrade lignin and hemicellulose, preserving cellulose [131]. Additionally, enzymes like xylanase and acetyl xylan esterase can be used to break down hemicellulose components, such as xylan [132].

Biological pretreatment offers several key advantages, including a reduced environmental impact, as it typically requires fewer chemicals and less energy compared to traditional methods. Enzymatic delignification improves the efficiency of subsequent processes, such as enzymatic hydrolysis, leading to higher biomass conversion rates. However, challenges remain, including the high cost of enzymes, slower processing times, and the need for specific enzymes tailored to different biomass types. To overcome these limitations, biological pretreatment is often combined with chemical methods to achieve more effective delignification and enhance overall process efficiency [128].

3.10. Comparison of Various Pretreatment Methods

Lignocellulosic biomass holds significant potential as a renewable resource for biofuel and biochemical production. However, its complex structure and inherent resistance to enzymatic degradation present substantial challenges to its efficient utilization [133]. To address these challenges, a range of pretreatment methods, both conventional and green, have been developed. Each method has distinct advantages and drawbacks, which influence its effectiveness in breaking down the biomass. Table 2 provides a comprehensive comparison of these pretreatment techniques, highlighting their respective strengths, limitations, and impacts on lignocellulosic biomass.

Table 2. Comparison between the various pretreatment methods.

Extraction method	Benefits	Limitations	Reference
Acid pretreatment	Maximized Cellulose Recovery Efficient Dissolution of Hemicellulose and Lignin	Potential for Equipment Corrosion Significant Waste Generation Requiring Post-Treatment High Expense Associated with Acid Reagents Adverse Environmental Consequences	[134]
Alkali pretreatment	Cost-Effective Efficient Lignin and Hemicellulose Solubilization Highly Effective for Delignification of Herbaceous Biomass	High Energy Consumption Corrosive Nature of Alkaline Agents Requirement for Neutralization Potential Partial Degradation of Hemicellulose and Cellulose	[135, 136]
Organosolv	High Purity of Cellulose Recyclability of Organic Solvents Efficient Recovery of Lignin and Hemicellulose for Value-Added Product Applications	High Energy Consumption Challenges in Recovering Organic Solvents Toxicity of Certain Organic Solvents Environmental Risks Associated with the Disposal of Organic Solvent Waste	[78]
Ionic Liquid	Strong Biomass Dissolution Capability Enhanced Cellulose Yields Environmentally Friendly Solvent Excellent Thermal Stability Low Vapor Pressure, Remarkable Tunability, and Synthetic Flexibility	High Volatility Increased Flammability Significant Toxicity Limited Biocompatibility and Poor Biodegradability	[137]
Biological pretreatment (Microorganism)	Mild Operating Conditions No Need for High Temperature or Pressure Low Energy Consumption Minimal or No Chemical Usage Negligible Environmental Impact	Slow Hydrolysis Rates Extended Pretreatment Duration Complexity in Microbial Synthesis Risk of Microbial Contamination Possible Need for Post-Treatment to Achieve High Cellulose Purity	[130]
Biological pretreatment (Enzymes)	Mild and Selective Process Environmentally Sustainable Minimal Inhibitor Production Low Energy Consumption Highly Specific and Eco-friendly Compatible with Biorefinery Integration Reduced Chemical Usage	High Enzyme Costs Limited Scalability Time-Intensive Process Requires Precise Control of Conditions Effectiveness Affected by the Recalcitrance of Certain Feedstocks Challenges in Enzyme Recovery	[138]
Supercritical fluid	Mild Pretreatment Conditions High Cellulose Yields Minimal Production of Fermentation Inhibitors Non-Toxic Nature of Supercritical Fluids	High Operational Costs Requirement for Specialized Equipment Potential Alteration of Cellulose Structure Inherent Safety Risks Due to High Pressure and Temperature Need for Advanced Understanding of Supercritical Fluid Properties (e.g., Phase Behavior, Cross-Over Region)	[139]
Deep Eutectic solvent	The solvent exhibits biodegradability, recyclability, low cost, wide availability, ease of preparation, low toxicity, and both chemical and thermal stability, enabling the selective solubilization of lignin at	The recovery of the solvent is hindered by the high viscosity of Deep Eutectic Solvents (DES). The range of suitable solvents is constrained. Scaling up the process presents significant chal-	[140]

Extraction method	Benefits	Limitations	Reference
Microwave-assisted treatment	mild temperatures. Deep Eutectic Solvents (DES) facilitate the preservation of cellulose within the biomass matrix. DES serves as a sustainable, environmentally benign solvent, consistent with green chemistry principles.	Challenges.	[105]
	Demonstrates high efficiency and rapid extraction performance. Enhances the properties of cellulose. Shortens treatment duration. Minimizes the need for chemicals and enzymes. Environmentally sustainable. Utilizes green pretreatment techniques.	Requirement for specialized equipment. Limited selectivity. Reactions occur at elevated temperatures. High capital investment and maintenance costs for the system. Reduced effectiveness with nonpolar or volatile solvents. Not suitable for processing large samples or agglomerated small samples. Extended cooling or venting times are necessary post-extraction.	
Ultrasound-Assisted Extraction	Decreased processing time. Minimal requirement for chemicals and enzymes. Sustainable and environmentally benign methods.	Low energy transfer efficiency within the vessel containing the extract. High equipment costs. Challenges in scalability due to limitations in equipment and processing capacity.	[141, 142]
Cold Atmospheric Plasma	Chemical-free processing. Versatile, rapid, cost-efficient, and environmentally sustainable. Reduced energy consumption. Enhanced reactivity of biomass.	Difficulties in controlling the reactive species in the plasma state. Restricted applicability to certain biomass types. Challenges associated with scaling up the process. High capital costs for equipment.	[143]
Steam explosion	The process requires no chemicals other than water. Capable of accommodating high biomass loadings. Capable of processing large particle sizes. Improved biomass digestibility.	Formation of toxic inhibitors that affect fermentation and enzymatic hydrolysis. Incomplete removal of lignin. Partial degradation of hemicellulose due to high pretreatment severities. Necessitate harsh processing conditions. Acid catalysts are required for biomass with high lignin content.	[86]
Hydrothermal processing	Accelerated reactions and rapid dissolution of biomass. Adjustable reaction conditions to meet specific requirements. Capability to dissolve a broad spectrum of lignocellulosic biomass. Minimal chemical usage. Elimination of the need for organic solvents. Reduction in the overall cost of the bio-conversion process.	Restricted to reactions under water-based or hydro-thermal conditions. Potential for corrosion-related issues. High energy consumption. Limited cellulose purity. Severe conditions are required to achieve effective pretreatment.	[144]

4. Cellulose Derivatives

Cellulose derivatives are a class of compounds synthesized by modifying the chemical structure of cellulose to impart specific properties or functionalities. This modification primarily involves altering the hydroxyl groups (OH) present on the glucose units [145], which are located at the C_2 , C_3 , and C_6 positions of the glucose rings. These hydroxyl groups serve as the key sites for chemical modification [145]. The derivatization of cellulose is achieved through chemical reactions with various reagents, resulting in the introduction of desired functional groups.

Two major categories of cellulose derivatives are cellulose ethers and cellulose esters [146]. The synthesis pathways for these derivatives are illustrated in Figure 2. Notable examples of cellulose ethers include methylcellulose (MC), ethyl cellulose (EC), hydroxyethyl cellulose (HEC), hydroxypropyl cellulose (HPC), hydroxypropyl methylcellulose (HPMC), carboxymethyl cellulose (CMC), and sodium carboxymethyl cellulose (NaCMC), most of which are water-soluble. In contrast, cellulose esters, such as cellulose acetate, cellulose acetate butyrate (CAB), cellulose nitrate, and cellulose propionate, tend to be insoluble in water [146].

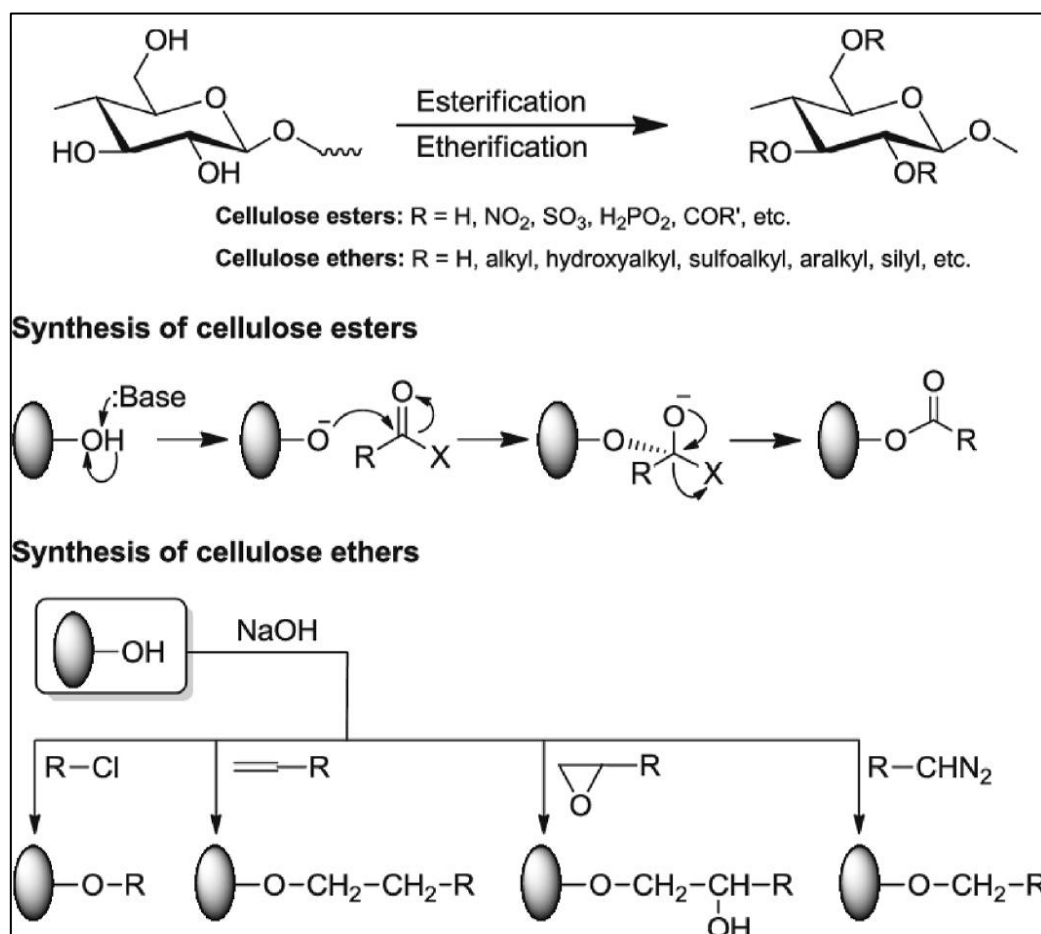


Figure 2. Synthesis pathways for cellulose esters and cellulose ethers [147].

4.1. Cellulose Ethers

4.1.1. Methylcellulose

Methylcellulose (MC) is a cellulose derivative synthesized by reacting alkali-treated cellulose with etherifying agents, such as dimethyl sulfate, iodomethane, or chloromethane. Following the reaction, MC undergoes a purification process, including washing and drying. The degree of substitution (DS), which refers to the number of hydroxyl groups replaced

by methoxyl groups in the cellulose structure, plays a crucial role in determining the solubility and industrial applications of MC [148, 149]. When the DS ranges from 0 to 3, MC is soluble in both water and organic solvents and exhibits distinct thermal properties. Below a critical temperature ($29 \pm 2^\circ\text{C}$), the viscosity remains stable or decreases, whereas, at higher temperatures, MC forms a thermoreversible gel [150].

MC is considered non-toxic, non-digestible, and non-allergenic, making it safe for consumption in food products worldwide. In the European Union, it is designated as E461 and is used as an emulsifier, texturizing agent in

baked goods, and stabilizer in frozen foods. In these applications, MC helps prevent the formation of fat crystals and ice crystals during freezing and thawing processes [151].

4.1.2. Carboxymethylcellulose

Carboxymethylcellulose (CMC) is an anionic, water-soluble cellulose derivative synthesized by reacting alkaline cellulose with chloroacetic acid or its sodium salt [152]. The synthesis involves two primary stages: mercerization, in which cellulose reacts with an alkali to form alkali-cellulose, followed by etherification, where sodium chloroacetate or chloroacetic acid is used to introduce carboxymethyl groups into the cellulose structure, resulting in the formation of CMC [37]. After the reaction, the mixture is filtered, neutralized, and subjected to purification steps including washing, filtration, and centrifugation, followed by drying to obtain the product in either a fine powder or granular form.

Unlike unmodified cellulose, CMC remains water-soluble at all temperatures due to the presence of ionic carboxymethyl groups. CMC's unique properties make it suitable for a wide range of applications across various industries, including detergents, paper manufacturing, adhesives, ceramics, cosmetics, food products, pharmaceuticals, textiles, and tissue engineering [153, 154].

4.1.3. Hydroxypropyl Methylcellulose

Hydroxypropyl methylcellulose (HPMC) is a synthetic cellulose derivative that appears as an odorless, tasteless, fibrous or granular white to off-white powder [155, 156]. It is synthesized by reacting partially depolymerized methylcellulose with hydroxypropyl groups, which enhance the viscosity, solubility, gelation, and film-forming characteristics of the cellulose structure [151]. Approved by both the FDA and the EU as a food additive, HPMC is widely utilized across multiple industries, including food, textiles, and healthcare, and has been investigated for use in biofilm packaging applications [157].

In the pharmaceutical sector, HPMC plays a critical role in the production of pills and tablets. Its high swellability is essential in controlled drug delivery systems, as it significantly influences the release kinetics of drugs, making it a valuable component in the design of sustained-release formulations [158, 159].

4.2. Cellulose Esters

Cellulose esters are synthesized by chemically modifying cellulose, replacing some of its hydroxyl groups with ester functional groups such as acetate, butyrate, or propionate. The process begins by swelling cellulose in a solvent, such as dimethylsulfoxide (DMSO) or acetone, to enhance the accessibility of its hydroxyl groups [160]. The activated cellulose is then reacted with an esterifying agent, such as acetic anhydride, in the presence of a catalyst (e.g., sulfuric acid or zinc chloride) at temperatures ranging from 60-80 °C to facili-

itate the esterification reaction.

Following the reaction, the mixture is purified by washing and filtering to remove impurities and excess reagents. Any remaining acid catalyst is neutralized with a base, such as sodium hydroxide. Common cellulose esters, including cellulose acetate, cellulose nitrate, and cellulose sulfate, are widely used in the production of coatings, films, and fibers due to their desirable properties [160, 161].

4.2.1. Cellulose Acetate

Cellulose acetate is a cellulose ester synthesized by substituting some of cellulose's hydroxyl groups with acetate ester functional groups. The production process begins by swelling cellulose in solvents such as dimethylsulfoxide (DMSO), which increases the accessibility of its hydroxyl groups. The activated cellulose is then reacted with an esterifying agent, such as acetic anhydride, in the presence of a catalyst, such as sulfuric acid or zinc chloride, at temperatures between 60-80 °C to promote the esterification reaction.

Following the reaction, the mixture is purified by washing and filtration to remove impurities and excess reagents. Any residual acid catalyst is neutralized with a base, such as sodium hydroxide. Cellulose acetate, along with other cellulose esters like cellulose nitrate and cellulose sulfate, is widely utilized in the production of coatings, films, and fibers due to its desirable properties in these applications [160, 161].

4.2.2. Cellulose Nitrate

Cellulose nitrate, commonly known as nitrocellulose or guncotton, is a highly flammable compound formed by reacting cellulose with concentrated nitric acid, which substitutes the hydroxyl groups with nitrate groups. Historically, nitrocellulose was primarily used as an explosive and propellant. It is soluble in a wide range of organic solvents, offering an alternative to more environmentally harmful polymers, such as polypropylene [160].

Cellulose nitrate has found applications in various products, including printing inks, wood coatings, nail lacquers, automotive paints, leather finishes, gun cotton, and flash paper. However, due to concerns regarding its flammability and potential instability over time, the use of cellulose nitrate has decreased. Safer alternatives are increasingly being developed and adopted in many of its previous applications.

4.3. Applications of Cellulose Derivatives

Cellulose derivatives are vital commercial products with diverse applications across various industries and household uses. These derivatives are incorporated into a wide array of products, such as coatings, laminations, optical films, and absorbent materials. Moreover, cellulose derivatives are utilized in the construction industry and serve as key additives in pharmaceutical, food, and cosmetic formulations. Table 3 provides a comprehensive overview of the production methods, applications, and commercially available cellulose-based

products, including their trade names and specific uses.

Table 3. Production and Applications of Cellulose Derivatives.

Derivative	Properties	Manufacturing process	Applications	References
Cellulose Acetate	Transparent, thermo-plastic, moisture-resistant, UV-stable, and capable of being molded and shaped.	The acetylation of cellulose is performed by reacting cellulose with acetic anhydride in the presence of a catalyst, typically sulfuric acid.	Eyewear frames, tool grips, cigarette filter components, and photographic films.	[162]
Cellulose Ether	A water-soluble, nonionic compound that functions as a thickening and stabilizing agent, emulsifier, and forms a gel upon heating.	The reaction of cellulose with alkylating agents, such as methyl chloride or ethylene oxide, in the presence of an alkaline catalyst.	Applications in pharmaceuticals, food products, oil drilling, personal care formulations, and construction.	[149, 157]
Carboxymethyl Cellulose (CMC)	A water-soluble, anionic, nontoxic compound that exhibits stability and strong water-binding capacity.	Alkaline treatment of cellulose followed by etherification using sodium monochloroacetate.	Applications in textile sizing, paper coatings, food additives, pharmaceuticals, cosmetics, and the removal of heavy metal ions.	[163, 164]
Hydroxyethyl Cellulose (HEC)	A water-soluble, nonionic compound that forms clear and flexible films.	Etherification of cellulose using ethylene oxide.	Applications in oil drilling, personal care formulations, construction materials, paints, textiles, and ceramics.	[162]
Microcrystalline Cellulose (MCC)	Exhibits superior binding and disintegrating properties, with a white, odorless, and tasteless appearance.	Controlled hydrolysis of cellulose in a concentrated acid medium to selectively remove the amorphous regions, followed by spray-drying.	Applications in pharmaceuticals (as a tablet filler and binder), food products, cosmetics, and plastics.	[164, 28]
Nitrocellulose	Highly flammable, soluble in alcohol and various solvents, and capable of forming a hard yet flexible film.	Treatment of cellulose with nitric acid, typically in the presence of sulfuric acid.	Applications in lacquers, automotive coatings, nail polish, early photographic film, explosives, rocket propellants, and contemporary gunpowder.	[149]
Cellulose Nanocrystals (CNCs)	Characterized by a high surface area, excellent mechanical strength, biocompatibility, and biodegradability.	Acid hydrolysis is employed to selectively remove the amorphous regions of cellulose.	Applications in composites, biomedical fields, electronics, and packaging materials.	[161-163]
Cellulose Nano fibres (CNFs)	Characterized by a high aspect ratio, exceptional mechanical strength, light weight, and flexibility.	Mechanical shearing or refining, typically in combination with enzymatic or chemical pretreatment processes.	Applications in composite materials, paper products, filtration systems, and textiles.	[27]
Carboxymethylcellulose (CMC)	Water-soluble, non-toxic, and biodegradable.	Cellulose is treated with sodium hydroxide, followed by reaction with monochloroacetic acid.	Applications in the food industry, pharmaceuticals, and cosmetics.	[163]
Hydroxypropyl cellulose (HPC)	Water-soluble, capable of forming clear solutions,	Cellulose is modified through treatment with propylene ox-	Applications as pharmaceutical excipients, coatings,	[158]

Derivative	Properties	Manufacturing process	Applications	References
	and thermoplastic in nature.	ide in the presence of a catalyst.	and adhesives.	
Ethyl cellulose (EC)	Insoluble in water, thermoplastic, and capable of forming flexible films.	Cellulose undergoes reaction with ethyl chloride in the presence of an alkali.	Applications as coatings, film-forming agents, and binders in pharmaceutical formulations.	[149]
Hydroxyethyl cellulose (HEC)	Water-soluble and capable of forming clear, viscous solutions.	Cellulose is modified through reaction with ethylene oxide under alkaline conditions.	Applications in paints, cosmetics, and detergents.	[163]
Methylcellulose (MC)	Water-soluble and capable of forming thermally reversible gels.	Cellulose is modified by treatment with methyl chloride in an alkaline solution.	Applications in the food industry, pharmaceuticals, and adhesive formulations.	[164, 149]

5. Conclusion and Future Directions

In conclusion, deep eutectic solvents (DES) and organosolv pretreatment methods emerge as highly efficient and environmentally friendly alternatives for cellulose extraction from lignocellulosic biomass, providing a promising pathway for sustainable biomass utilization. These methods are effective in dissolving lignin and hemicellulose while maintaining cellulose integrity, thereby facilitating its conversion into valuable products. Among traditional pretreatment strategies, mild alkaline treatment continues to be the most widely used, owing to its demonstrated effectiveness in removing lignin and hemicellulose, thus enhancing cellulose accessibility for subsequent processing.

Agricultural residues, which are abundant, renewable, and cost-effective, serve as an essential feedstock for cellulose extraction, aligning with the principles of a circular economy and sustainability. Future research should focus on optimizing and scaling up fractionation technologies to ensure high-quality cellulose production while maintaining or improving yield. Addressing the challenges associated with extraction methods, such as cost, efficiency, and scalability, will be critical for realizing the full potential of lignocellulosic biomass as a sustainable resource. This will, in turn, advance the development of green, economically viable solutions for the bio-based economy.

Abbreviations

LCB	Lignocellulosic Biomass
NaOH	Sodium Hydroxide
SE	Steam Explosion
HMF	Hydroxymethylfurfural
HPP	High-Pressure Processing
HHP	High Hydrostatic Pressure
UHP	Ultrahigh-Pressure Processing
DESs	Deep Eutectic Solvents

HBD	Hydrogen Bond Donor
SFE	Supercritical Fluid Extraction
CO ₂	Carbon Dioxide
WRF	White-Rot Fungi
SSF	Solid-State Fermentation
OH	Hydroxyl Groups
MC	Methylcellulose
EC	Ethyl Cellulose
HEC	Hydroxyethyl Cellulose
HPC	Hydroxypropyl Cellulose
HPMC	Hydroxypropyl Methylcellulose
CMC	Carboxymethyl Cellulose
NaCMC	Sodium Carboxymethyl Cellulose
CAB	Cellulose Acetate Butyrate
DS	Degree of Substitution
DMSO	Dimethylsulfoxide

Conflicts of Interest

The authors declare no conflicts of interest.

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