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Review

Physical–Chemical–Biological Pretreatment for Biomass Degradation and Industrial Applications: A Review

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Abstract: Lignocellulosic biomass, including agricultural, forestry, and energy crop waste, is one of Earth's most abundant renewable resources, accounting for approximately 50% of global renewable resources. It contains cellulose, hemicellulose, and lignin, making it crucial for biofuels and bio-based chemicals. Due to its complex structure, single-pretreatment methods are inefficient, leading to the development of combined pretreatment technologies. These methods enhance cellulose accessibility and conversion efficiency. This paper analyzes the principles, advantages, and disadvantages of various combined pretreatment methods and their practical benefits. It highlights recent research achievements and applications in biofuel, biochemical production, and feed. By integrating multiple pretreatment methods, biomass degradation efficiency can be significantly improved, energy consumption reduced, and chemical reagent use minimized. Future advancements in combined physical, chemical, and biological pretreatment technologies will further enhance biomass utilization efficiency, reduce energy consumption, and protect the environment, providing robust support for sustainable renewable energy development and ecological protection.

Keywords: lignocellulose; combined pretreatment; physical–chemical–biological; biochemical products; sustainable renewable energy

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1. Introduction

Lignocellulose is primarily composed of cellulose, hemicellulose, and lignin, forming a complex and robust network structure [1]. Cellulose is a polymer composed of glucose units linked by β-1,4-glycosidic bonds [2], hemicellulose is a heteropolysaccharide made up of various sugars (such as xylose, arabinose, and glucose) [3], and lignin is a macromolecule formed by aromatic monomers (such as guaiacyl and syringyl units) connected through phenylpropane units [4]. These components intertwine to create the recalcitrance and resistance to degradation of lignocellulose (Figure 1). The high crystallinity of cellulose and the barrier effect of lignin make lignocellulose difficult to degrade and utilize, significantly limiting its efficiency and economic viability in biorefining processes [5]. Table 1 shows the lignocellulose content in various types of biomass resources.

To improve the utilization of lignocellulose, pretreatment is typically required. Traditional pretreatment methods include physical pretreatment [6], chemical pretreatment [7], and biological pretreatment [8], each of which has its own limitations. Single-pretreatment methods often face issues such as low efficiency, high by-product formation, and high costs when dealing with lignocellulose [9]. For example, although acid pretreatment can effectively remove hemicellulose, it may produce inhibitory by-products such as furfural and 5-hydroxymethylfurfural (HMF). These by-products can inhibit subsequent microbial fermentation processes, affecting the yield and quality of the products [10]. Alkaline pretreatment can effectively remove lignin and improve cellulose accessibility, but it requires harsh conditions, such as high temperature or high pressure [11]. Additionally, this process typically necessitates subsequent neutralization treatment to remove residual alkaline substances, thereby increasing the cost and complexity of the pretreatment. Combined pretreatment refers to the use of a combination of physical, chemical, and biological methods to process biomass, with the goal of enhancing its degradability and conversion efficiency [12,13]. To overcome the limitations of single-pretreatment methods, combining multiple pretreatment approaches allows for the complementary advantages of different methods, thereby improving the degradation efficiency and utilization of lignocellulose [14]. This approach has significant applications in fields such as biorefining, wastewater treatment, and biodegradation.

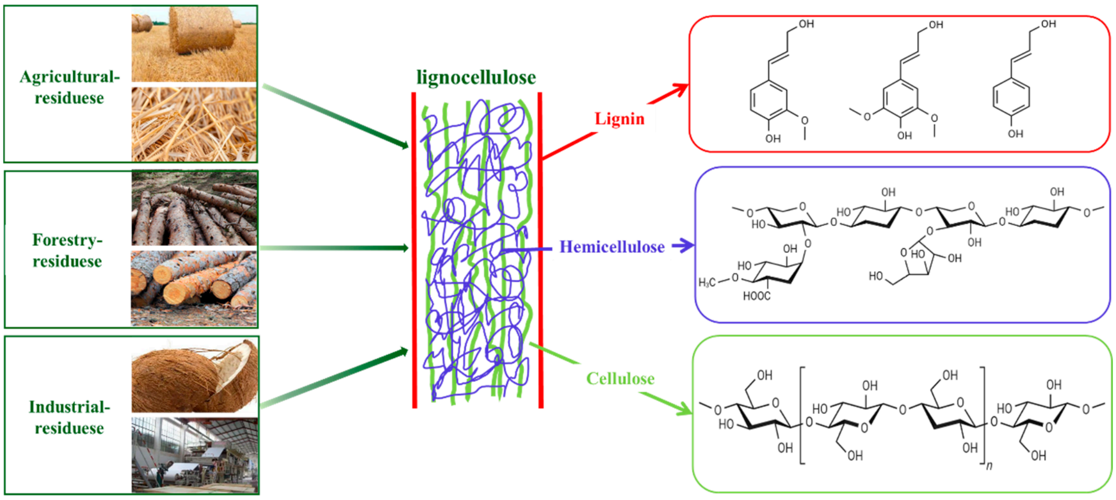

Figure 1. Structural arrangement and composition of lignocellulosic biomass.

Combined pretreatment methods, through the synergistic action of multiple techniques, can more efficiently disrupt the structure of lignocellulose, exposing cellulose and hemicellulose. This significantly enhances the accessibility of cellulose and hemicellulose, making subsequent enzymatic hydrolysis and fermentation processes more efficient [15] (Figure 2). This approach not only improves the sugar conversion rate but also reduces the energy consumption and cost of pretreatment. Yang et al. [16] demonstrated that combined wet alkali and mechanical pretreatment of corn straw removed 44.4% of lignin while retaining a significant portion of cellulose (86.6%). This method offers the advantages of a shorter processing time and reduced chemical consumption. The combined pretreatment using physical, chemical, and biological methods can reduce the crystallinity of cellulose, enhance enzyme accessibility, and thereby increase the conversion rate of cellulose [14,17]. Combined pretreatment can optimize the use of chemical reagents and reduce the formation of toxic by-products. For instance, Dziekońska-Kubczak et al. [18] used a combination of acid (HNO_3) and alkali (NaOH) for pretreating Jerusalem artichoke stalks (JAS) and oat straw (OS). They first applied 5% nitric acid followed by NaOH pretreatment, achieving the highest glucose yields with enzymatic hydrolysis efficiencies of 90.6% for JAS and 97.6% for OS. Acid–alkali combined pretreatment can lower the amounts of acid or alkali used individually, reduce the burden of waste liquid treatment, and decrease the risk of environmental pollution. The utilization efficiency of lignocellulose is significantly improved through combined pretreatment technologies, providing a more economical, environmentally friendly, and efficient solution for biorefining. This advancement promotes the sustainable development of biomass energy and bio-based products.

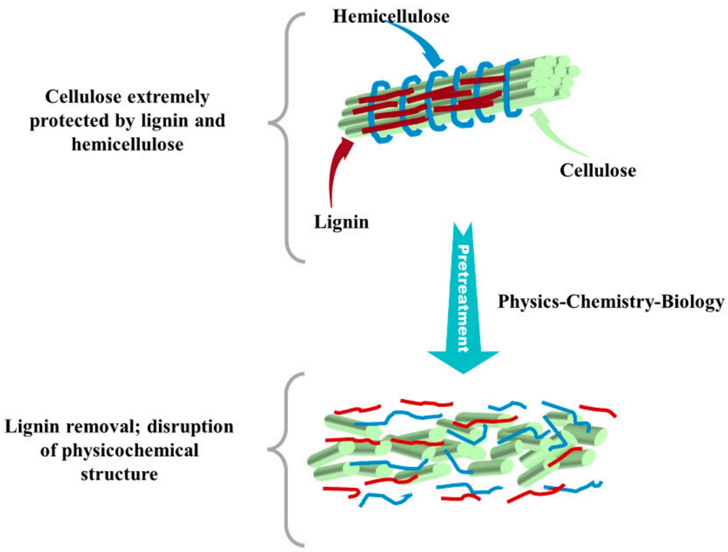

Figure 2. Functions of pretreatment.

Lignocellulose is an abundant renewable resource, and through efficient combined pretreatment technologies, it can be effectively converted into biofuels (such as ethanol and methane) [8,19] and bio-based chemicals (such as organic acids and platform chemicals) [20,21], thus reducing dependence on fossil fuels and advancing the development of renewable energy. Combined pretreatment not only improves the degradation efficiency of lignocellulose but also facilitates the development of various high-value products, including bio-materials [22,23], animal feed [24], and fertilizers [25]. This helps to build a diversified biorefining industry chain and enhances economic benefits. Additionally, this conversion process promotes the resource utilization of agricultural and forestry waste, advancing the development of a circular economy. Therefore, researching and optimizing pretreatment and conversion technologies for lignocellulosic biomass is significant not only in the fields of energy, chemicals, and materials but also in achieving comprehensive environmental and social benefits, thus promoting the sustainable development of renewable energy, green chemistry, and bio-material industries.

Table 1. Chemical composition of various biomass resources (% dry basis).

Category	Material	Cellulose (%)	Hemicellulose (%)	Lignin (%)	References
Agricultural residues	Corn stover	28–40%	25–35%	10–20%	[26]
	Wheat straw	35–40%	20–30%	15–20%	[26]
	Rice straw	32–47%	19–27%	5–24%	[27]
Forestry residues	Hardwood (Birch)	40–45%	25–35%	20–25%	[28]
	Softwood (Pine)	40–44%	25–29%	26–35%	[29]
Industrial residues	Bagasse	32–44%	27–32%	19–24%	[30]
	Paper mill sludge	30–50%	50–15%	5–10%	[31]
Dedicated energy crops	Willow	42–49%	16–20%	23–25%	[32]
	Sweet sorghum	30–40%	25–30%	10–15%	[33]
Aquatic plants	Water hyacinth	17–21%	35–45%	15–17%	[34]
	Algae	~70%	~43%	NA	[35]
Fruit shells and pomace	Coconut shell	26–35%	15–20%	29–36%	[36]
	Olive pomace	25–35%	20–25%	30–35%	[37]

This review aims to explore the application and advantages of combined pretreatment technologies in the conversion of lignocellulosic biomass. Lignocellulosic biomass, one of the most abundant renewable resources on Earth, mainly includes agricultural waste, forestry residues, and energy crops. Due to its complex structure, single-pretreatment methods are often insufficient for efficient degradation, leading to the emergence of combined pretreatment technologies. This paper provides a detailed analysis of the principles, advantages, and disadvantages of various combined pretreatment methods, as well as their application potential in the production of biofuels, bio-based chemicals, and feed and fertilizers. By integrating multiple pretreatment methods, it is possible to significantly enhance biomass degradation efficiency, reduce energy consumption, and decrease the use of chemical reagents, thereby promoting the sustainable development of renewable energy and green chemicals. In the future, combined pretreatment technologies involving physical, chemical, and biological methods will continue to improve and play a greater role in the efficient utilization of biomass resources, reduction in energy consumption, and environmental protection, providing strong technical support for achieving sustainable development.

2. Combined Pretreatment

Single-pretreatment technologies (e.g., physical, chemical or biological pretreatment) have shown some success in the degradation of lignocellulosic biomass [38,39]. However, these single-pretreatment methods have some significant limitations in practice, such as low conversion efficiency, high energy or chemical requirements, and limited applicability [40]. In view of the limitations of single-pretreatment methods, combined pretreatment technology has become an important development direction to enhance the conversion efficiency of lignocellulosic biomass. Combined pretreatment refers to the use of multiple pretreatment methods in combination when processing lignocellulosic biomass (such as wood and crop straw) [15,41]. The aim is to break down the complex structure of the biomass, increase the accessibility of cellulose, and thereby improve the efficiency of subsequent processes (such as enzymatic hydrolysis and fermentation) [42,43] (Figure 3). Combined pretreatment technology has obvious advantages such as synergistic effect and wider applicability [44,45]. Therefore, it is necessary and urgent to conduct systematic studies on combined pretreatment technologies. This review synthesizes and various combined pretreatment, discusses its pretreatment process and effect, and provides theoretical basis and technical guidance for the efficient conversion of biomass resources.

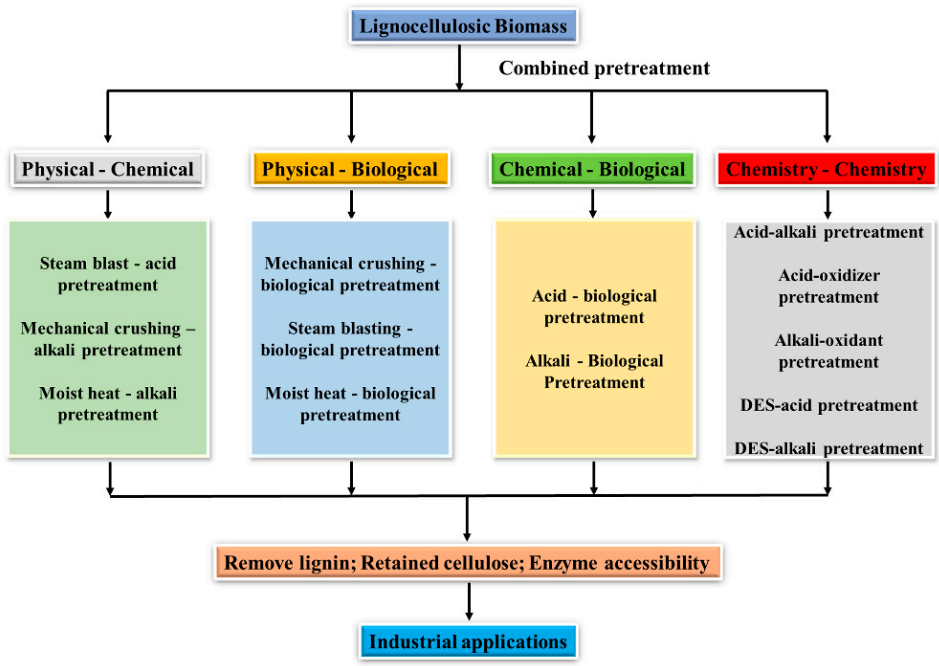

Figure 3. Types of combined pretreatment for lignocellulosic biomass resources.

2.1. Physical–Chemical Combined Pretreatment

Physical–chemical combined pretreatment is a technique that integrates physical methods with chemical methods for the pretreatment of lignocellulosic biomass [14]. This approach aims to use physical means to enhance the efficiency of chemical pretreatment, or to utilize chemical methods to boost the effectiveness of physical pretreatment, achieving better degradation and conversion outcomes.

2.1.1. Steam Explosion and Chemical Pretreatment

Steam explosion (SE) is a clean technology used for the pretreatment of lignocellulosic biomass, using only water instead of chemicals, and is therefore considered to have a minimal environmental impact [46]. Biomass materials are placed in a reactor, injected with high-temperature and high-pressure steam (typically 160–260 °C, pressure of 0.69–4.83 MPa). After a certain period of steam treatment (from seconds to minutes), the pressure is rapidly released, causing partial hydrolysis of the hemicellulose portion of the lignocellulosic material, while partially breaking down the lignin structure, thus increasing the accessibility of cellulose. Semwal et al. [47] crushed rice straw to 5–20 mm and found that, after dilute acid soaking and subsequent high-temperature steam explosion pretreatment, the hemicellulose content decreased to 5.7–7.4%, and the cellulose enzymatic hydrolysis rate reached 88.7–89.6%. First, steam explosion pretreatment exposes the lignocellulose to a high-temperature, high-pressure steam environment, and a rapid pressure drop causes the cellulose structure to burst, increasing its specific surface area. Then, acid pretreatment allows the acid to more effectively hydrolyze cellulose and hemicellulose [48]. Sui et al. [49] proposed a combined method of steam explosion without adding any acid or base catalyst followed by room temperature γ-valerolactone (GVL)/water delignification to enhance the enzymatic saccharification of corn stover. After steam explosion at 1.5 MPa (198 °C) for 10 min, up to 70% of the original xylan was removed from the corn stover, while retaining over 92% of the glucan. Combining urea and steam explosion pretreatment of corn stover, with a urea addition rate of 4.87% and steam pressure of 1.22 MPa, yielded a maximum reducing sugar of 350.12 mg/g and a maximum sugar alcohol conversion rate of 48.3% [50]. Katsimpouras et al. [51] combined acid and steam explosion pretreatment of corn stover and found that after 0.2% H_2SO_4 combined with steam explosion, hemicellulose content decreased to 7.8%. However, steam explosion requires high-end equipment, complex operations, and high energy consumption. Further optimization of steam explosion and acid treatment process parameters, such as temperature, pressure, acid concentration, and treatment time, is necessary to achieve more efficient and cost-effective pretreatment.

2.1.2. Mechanical Crushing and Chemical Pretreatment

Mechanical crushing breaks down lignocellulosic raw materials into small particles, increasing their specific surface area and porosity [6,8]. This is followed by chemical pretreatment (such as acid, alkali, or oxidizing agent pretreatment) to allow the chemical reagents to act more uniformly and effectively on cellulose, hemicellulose, and lignin. Yu, et al. [6] found that the combined method of mechanical crushing and acid pretreatment resulted in more than doubling the enzymatic hydrolysis efficiency of cellulose compared to mechanical crushing alone. Corn stover was ground and sieved to 30–50 mesh, and under conditions of 110 °C, 4% NaOH, 90 min reaction time, and 60% (*v*/*v*) ethanol, the total lignin removal rate exceeded 80%, with minimal degradation of hemicellulose. After enzymatic hydrolysis, the maximum total monosaccharide recovery rate was 83.7% (cellulose 85.0%, hemicellulose 82.0%) [52]. Mechanical crushing technology overcomes the recalcitrant structure by breaking down the physical barrier of the cell wall, reducing cellulose crystallinity, and removing lignin, allowing hydrolytic enzymes to access the biomass macrostructure. Yang, et al. [16] used 3% NaOH combined with ball milling technology to pretreat corn stover, removing 44.4% of the lignin and achieving a cellulose enzymatic hydrolysis rate of 91.3%. While mechanical crushing can enhance the efficiency of chemical pretreatment and reduce the amount of chemical reagents needed, it is energy-intensive and may require high-energy consumption equipment.

2.1.3. Moist Heat–Alkali Pretreatment

Moist heat pretreatment involves processing lignocellulose in a high-temperature, high-pressure steam environment to loosen its structure and partially remove lignin. This is followed by alkali pretreatment, where the alkali solution can penetrate deeper into the cellulose and hemicellulose, enhancing the dissolution and removal effects. Hydrothermal pretreatment of wheat straw indicated that 60 min is the optimal pretreatment time to achieve the highest substrate dissolution [53]. Romaní et al. [54] used 0.4 g/g substrate of lime at 121 °C for 1 h to pretreat oat straw, achieving a lignin removal rate of 57%, cellulose enzymatic hydrolysis rate of 99%, and maximum ethanol yield of 50 g/L. Rapeseed straw subjected to hydrothermal pretreatment followed by alkali pretreatment showed that the solid recovery rate dropped to below 50%, with lignin and hemicellulose content reduced to below 10% and 5%, respectively [55]. Moist heat pretreatment of lignocellulose can enhance the efficiency of alkali pretreatment, reduce treatment time, and lower alkali consumption.

In summary, Physical–chemical combined pretreatment demonstrates significant technological advantages and market potential in the utilization of biomass resources, providing crucial technical support and development directions for achieving sustainable development and innovative economies.

2.2. Physical–Biological Combined Pretreatment

Physical–biological combined pretreatment is a lignocellulosic pretreatment technology that integrates physical and biological methods. Its aim is to enhance the effectiveness of biological pretreatment through physical means and to improve the efficiency of physical pretreatment by combining it with biological approaches.

2.2.1. Mechanical Pulverization–Biological Pretreatment

Mechanical pulverization crushes lignocellulosic materials into smaller particles, increasing their specific surface area and porosity, which facilitates the penetration and degradation of cellulose, hemicellulose, and lignin by microorganisms or enzymes. Subsequently, biological pretreatment is carried out, such as using white rot fungi or cellulases for treatment. Pulverizing poplar bark to 0.4 mm and applying white rot fungi (e.g., *Ceriporiopsis subvermispora*, *Coprinus cinereus*, and *Populus ostreatus*) for biological pretreatment can achieve a lignin degradation rate of over 50%, outperforming any single fungal strain treatment [56]. After grinding the straw to 0.45 mm and composting it with mixed microorganisms for 14 days, the degradation rates of cellulose, hemicellulose, and lignin were 44.4%, 34.9%, and 39.2%, respectively. This pretreatment is advantageous for increasing methane production during the anaerobic fermentation of corn stover with mixed microorganisms and for shortening the fermentation cycle [57]. Microbial combined with dilute acid pretreatment is a promising method for water hyacinth hydrolysis. Pulverizing water hyacinth to 40 mesh and applying *Phanerochaete chrysosporium* for biological pretreatment results in a cellulose content of 39.4% and a reducing sugar yield of 430.66 mg/g, without the addition of any extra cellulase [34]. Mechanical pulverization of lignocellulosic materials can enhance the efficiency of biological pretreatment and reduce processing time.

2.2.2. Steam Explosion–Biological Pretreatment

Biomass treated with steam explosion (SE) becomes more amenable to enzymatic hydrolysis, significantly improving sugar release efficiency. Subsequent biological pretreatment allows microorganisms or enzymes to more effectively degrade cellulose and hemicellulose. Shi et al. [58] reported that after SE pretreatment of corn stover, hemicellulose was reduced by 28.00%, and further co-fermentation with anaerobic fungi and methanogens led to an additional 12.8% reduction in hemicellulose. SE is often regarded as one of the most cost-effective pretreatment technologies. For example, using SE pretreatment to enhance the biochemical methane potential of Miscanthus increased the potential by up to 51.3%, with scanning electron microscopy (SEM) images showing significant disruption of the recalcitrant structure of *Miscanthus lutarioriparius* [59]. SE pretreatment can improve the efficiency of biological pretreatment and shorten processing time, but it requires high equipment standards, complex operation, and substantial energy consumption. Future research will focus on optimizing SE process parameters (such as temperature, pressure, and treatment time) to further enhance pretreatment efficiency and reduce energy consumption. Combining SE with other pretreatment methods (such as alkaline peroxide treatment and ionic liquid treatment) [60,61] could achieve more efficient lignocellulose degradation. Advancing SE technology for industrial-scale applications and developing large-scale pretreatment devices and systems will improve biomass conversion efficiency and economic benefits.

2.2.3. Hydrothermal–Biological Pretreatment

Hydrothermal pretreatment involves treating biomass with high-temperature water or steam to hydrolyze hemicellulose and partially depolymerize lignin, thereby increasing the accessibility of cellulose. After treatment, the biomass is rapidly cooled to stabilize the structural changes [62]. During this process, when the temperature exceeds 170 °C, acetic acid is formed from the acetyl groups present in hemicellulose, catalyzing the hydrolysis reaction [63]. The presence of acetic acid further loosens the biomass structure by removing additional hemicellulose, which enhances the convertibility of cellulose. Subsequently, biological pretreatment allows microorganisms or enzymes to more effectively degrade cellulose and hemicellulose, improving degradation efficiency. For instance, hydrothermal pretreatment of safflower straw under optimal conditions (120 °C for 1 h) resulted in 148.4 m^3 of methane per ton of treated straw, compared to 86.9 m^3 of methane from untreated straw. Additionally, enzymatic hydrolysis of the solid fraction showed that under the most severe pretreatment conditions (180 °C for 5 h) with an enzyme loading of 10 FPU/g substrate, the highest released sugar concentration was 25.1 g/L, compared to 4.5 g/L from untreated biomass [64]. Song et al. [65] studied the pretreatment characteristics and anaerobic digestion (AD) performance of corn stover under different severities of hydrothermal pretreatment. The highest removal rates of hemicellulose and lignin were 95.41% and 13.85%, respectively, at severities of 6.81 and 1.98.

Hydrothermal pretreatment significantly enhances biomass accessibility, and subsequent biological treatment further degrades hemicellulose, improving sugar release rates. Future research should focus on waste management and resource recovery technologies in the hydrothermal–biological combined pretreatment process to reduce environmental impact and increase overall process sustainability.

2.3. Chemical–Biological Combined Pretreatment

Chemical–biological combined pretreatment is a lignocellulosic pretreatment technique that integrates chemical and biological methods. The chemical pretreatment initially disrupts the biomass structure, enhancing the effectiveness of subsequent biological treatment. Following this, biological pretreatment utilizes microorganisms or enzymes to further degrade cellulose and hemicellulose, producing fermentable sugars [66]. Chemical pretreatment significantly increases the accessibility of biomass, while biological treatment further degrades cellulose and hemicellulose, improving sugar release rates. By optimizing chemical pretreatment conditions and integrating biological pretreatment, the formation of inhibitors (such as furfural and HMF) can be minimized, thus enhancing fermentation efficiency. This approach is suitable for various lignocellulosic biomasses, including crop residues, wood chips, and herbaceous plants.

2.3.1. Acid Pretreatment–Biological Pretreatment

Acid pretreatment is first conducted using dilute acids (such as dilute sulfuric acid or hydrochloric acid) to hydrolyze hemicellulose and partially cellulose, disrupting the lignocellulosic structure and making it more porous. Subsequently, biological pretreatment is performed, utilizing microorganisms or enzymes to further degrade cellulose and lignin. Martínez Patiño, et al. [44] combined fungal pretreatment with chemical pretreatment and found that the order of the pretreatment steps significantly affects glucose yield. The optimal approach was to first perform fungal pretreatment with *Irpex lacteus* for 28 days, followed by dilute acid pretreatment (2% *w*/*v* H_2SO_4, 130 °C, 90 min), which resulted in a 34% increase in enzymatic hydrolysis yield compared to acid pretreatment alone. Similarly, *Phanerochaete chrysosporium* was used for biological pretreatment of water hyacinth ground to 40 mesh, followed by acid pretreatment with 1% H_2SO_4 at 100 °C. This combination reduced lignin content from 0.43 g to 0.13 g and increased the yield of reducing sugars to 430.66 mg/g [34]. Acid pretreatment significantly enhances the accessibility of cellulose, improving the efficiency of biological pretreatment. However, acid pretreatment may produce toxic by-products that require neutralization, increasing process complexity and cost.

2.3.2. Alkaline–Biological Pretreatment

Alkaline pretreatment is an effective and cost-efficient method for producing fermentable sugars, primarily using alkaline solutions such as sodium hydroxide, potassium hydroxide, or calcium hydroxide to treat biomass [67,68]. Alkaline solutions can disrupt the lignin structure, dissolve some hemicellulose, and increase the accessibility of cellulose. By breaking the chemical bonds in lignin and hemicellulose, the surface area of exposed cellulose is increased, enhancing the efficiency of subsequent biological pretreatment. Alkaline pretreatment partially degrades lignin and hemicellulose, which reduces the amount of enzyme required during biological pretreatment, thus lowering enzyme costs. Key factors affecting lignin removal and fermentable sugar production include alkaline load, reaction time, and temperature [69]. For example, water hyacinth ground to 40 mesh was first biologically pretreated with *Phanerochaete chrysosporium* and then subjected to alkaline pretreatment with 4% NaOH at 100 °C. This process reduced the lignin content from 0.43 g to 0.12 g, significantly removing most of the lignin, increasing enzyme accessibility, and raising the yield of reducing sugars to 430.66 mg/g [34]. Although biological pretreatment has the advantages of environmental friendliness and low energy consumption, it typically requires a longer pretreatment time. Zhong et al. [70] investigated a novel approach combining white-rot fungi and alkaline pretreatment at near-room temperature for the saccharification of corn stover to accelerate the biological process. Biological pretreatment with *Irpex lacteus* or *Echinodontium taxodii* significantly improved the enzymatic hydrolysis of corn stover, but the process required a long time (60 d) to achieve satisfactory sugar yields. However, when biological pretreatment was combined with alkaline pretreatment, the biological process time was reduced to 15 d, and the efficiency of alkaline pretreatment was significantly improved. The final glucose yield from combined pretreatment was 271.1 mg/g, which was a 50.4% and 28.3% increase compared to single-alkaline pretreatment under the same and optimal reaction times, respectively.

Compared to acid pretreatment, alkaline pretreatment generates fewer inhibitors (such as furfural and HMF), which is beneficial for the subsequent fermentation process [71]. By optimizing pretreatment conditions, the generation of inhibitors can be further reduced, and fermentation efficiency improved. Alkaline–biological combined pretreatment can reduce overall pretreatment time and energy consumption, enhancing economic benefits. Comprehensive utilization of by-products enables full component utilization of biomass, further improving economic viability and sustainability.

2.4. Chemical–Chemical Combined Pretreatment

Chemical–chemical combined pretreatment refers to the use of two or more chemical methods in conjunction to disrupt the structure of lignocellulose, thereby enhancing its subsequent enzymatic hydrolysis or fermentation efficiency. This combined pretreatment approach leverages the advantages of different chemical methods to overcome the limitations of a single method, achieving superior pretreatment results.

2.4.1. Acid–Alkali Pretreatment

Acid–alkali combined pretreatment involves using both acidic (such as sulfuric acid or hydrochloric acid) and alkaline methods to treat biomass. The acidic treatment disrupts the chemical bonds in hemicellulose and lignin, generating low-molecular-weight compounds that make cellulose more accessible for subsequent processing [72,73]. The alkaline treatment primarily targets lignin, dissolving some hemicellulose and increasing the accessibility of cellulose. This combined approach significantly enhances biomass accessibility and degradation efficiency, with acidic pretreatment focusing on hemicellulose and alkaline pretreatment on lignin, resulting in a complementary and effective process.

Under conditions of 1.81% (w/v) NaOH concentration and a solid-to-liquid ratio of 5 (w/v), alkaline hydrolysis of corn cobs for 90 min achieved a lignin removal rate of 82.03%. Subsequent pretreatment with 6% (w/v) H_2SO_4 resulted in a xylose yield of 74% [74]. Li et al. [75] employed a two-stage acid/alkali pretreatment for sorghum stalks, demonstrating better saccharification performance compared to conventional single-stage pretreatment. The acid–alkali pretreatment achieved a higher glucose yield (0.23 g/g), which is 1.64 times and 1.21 times greater than single-stage pretreatment and acid–alkali pretreatment, respectively. The acid–alkali combined pretreatment process is a viable method for achieving high fermentation glucose conversion rates in cellulose materials. It is well-known that acidic pretreatment can extract hemicellulose in the form of pentoses and some lignin, producing biomass rich in cellulose and lignin. The liquid fraction obtained from acid treatment, which is rich in pentoses, can be fermented using pentose fermenting microbes (*Pichia stipitis* and *Pichia pastoris*). Kaur and Kuhad [76] developed a bioprocess utilizing all biopolymers in lignocellulosic rice straw. The biomass was first acid-pretreated and then alkali-pretreated to separately target the removal of hemicellulose and lignin. Acid treatment removed 90% of hemicellulose, resulting in an acid hydrolysis product with a monomeric sugar concentration of 20 g/L. Alkali treatment removed 55% of lignin, and the resulting biomass had a total cellulose content of 830 mg/g. Enzymatic saccharification of the pretreated biomass produced 787 mg/g of reducing sugars, with hexose and pentose fermentation yields of 0.40 and 0.47 g/g, respectively. Chen et al. [77] mixed acid and alkali pretreated materials while controlling the pH without washing, which produced more ethanol (19.2 g/L) compared to separate acid or alkali pretreatments.

This method is applicable to various types of lignocellulosic biomass, such as crop residues, wood chips, and herbaceous plants, offering strong versatility. By properly controlling the amount of acid and alkali and the processing conditions, environmental pollution and waste treatment needs can be minimized, increasing the overall sustainability of the process.

2.4.2. Acid–Oxidant Pretreatment

Acid–oxidant combined pretreatment involves using an oxidant to treat biomass, which disrupts the aromatic ring structures in lignin and partially degrades hemicellulose, thus increasing the accessibility of cellulose. This combined approach leverages the synergistic effects of acids and oxidants to enhance the degradation efficiency of lignocellulosic biomass. Acidic pretreatment primarily targets hemicellulose, while oxidant pretreatment focuses on lignin, resulting in a complementary and highly effective process. Wang et al. [78] used a phosphoric acid and hydrogen peroxide (PHP) pretreatment on wheat straw. The results showed that almost all xylan was removed, along with over 70% of the lignin, with more than 90% of the cellulose recovered in the solid fraction. This method is versatile and can be applied to various feedstocks, including softwoods, hardwoods, agricultural residues, bamboo, garden waste, and their mixtures, achieving almost complete removal of hemicellulose and 70–100% removal of lignin [79]. Pretreatment of wheat straw with phosphoric acid and hydrogen peroxide can enhance enzymatic saccharification and lignin removal, increasing cellulose content to 68.9%, with nearly 100% saccharification rate [80]. An, et al. [73] employed a two-stage dilute sulfuric acid (DA) and ammonium water wet oxidation (AWO) pretreatment to recover sugars from corn stalks. In the first stage, at 120 °C, 40 min, and 1 wt% HCl, 82.8% of xylan was recovered. The second stage, conducted under milder conditions (130 °C, 12.6 wt% ammonium hydroxide, 3.0 MPa O_2, 40 min), removed 86.1% of lignin.

Oxidants effectively disrupt lignin structures, significantly improving cellulose accessibility and increasing the efficiency of subsequent biological processing. Compared to acid pretreatment, oxidant pretreatment generates fewer inhibitory by-products, which benefits the subsequent fermentation process.

2.4.3. Alkaline–Oxidant Pretreatment

Alkaline (e.g., NaOH) and alkaline peroxide pretreatments are effective chemical methods for treating lignocellulosic materials. Hydrogen peroxide, under acidic or alkaline conditions, decomposes to produce highly oxidative hydroxyl radicals (-OH). These radicals can effectively disrupt the aromatic ring structures of lignin [81]. The radicals attack the aromatic rings of lignin, causing them to cleave and generate low-molecular-weight aromatic compounds. Hemicellulose is also partially degraded, increasing the exposure of cellulose. Cao et al. [82] used a 2% (*w*/*v*) sodium hydroxide solution combined with 5% (*w*/*v*) hydrogen peroxide for immersion pretreatment. This method improved the removal rates of hemicellulose and lignin from sweet sorghum bagasse and enhanced cellulose retention, resulting in a glucose concentration in the hydrolysate of 14.16 mg/mL, which is 9.8 times higher than the control group. Chen et al. [83] employed a Na_2CO_3-O_2 combined pretreatment on wheat straw at 110 °C, achieving a lignin removal rate of 42% and a cellulose saccharification efficiency of 66.1%. Alkaline hydrogen peroxide pretreatment reduced the lignin content in bagasse, with a maximum lignin removal of 89 ± 3% (*w*/*w*) at 50 °C and 150 min. The effectiveness of lignin removal was confirmed by scanning electron microscopy, X-ray diffraction, and Fourier-transform infrared spectroscopy [84]. The use of dilute acid and alkaline H_2O_2 pretreatment shows promise for reducing process costs and making large-scale applications feasible. Alkaline H_2O_2 pretreatment achieved a glucose concentration of 62.4 g/L and removed 56.8% of lignin, resulting in a 71% glucose yield [85].

The synergistic action of alkali and oxidants can more efficiently remove lignin and hemicellulose, enhancing cellulose enzymatic hydrolysis efficiency. However, the process is complex, oxidants are costly, and the treatment may produce toxic oxidative by-products.

2.4.4. Deep Eutectic Solvent–Acid Pretreatment

Deep eutectic solvents (DESs) are mixtures composed of two or more substances that form a liquid through hydrogen bonding or van der Waals forces. Abbott et al. first described DESs as potential alternative solvents to ionic liquids (ILs) [86]. DESs are emerging due to their variety, design flexibility, low cost, green nature, high adjustability, ease of synthesis, recyclability, high solubility, biocompatibility, biodegradability, non-flammability, environmental friendliness, and 100% atomic economy [87]. DESs can dissolve cellulose and some lignin in biomass, disrupting their crystalline structure and increasing cellulose accessibility. Following this, acid pretreatment can more effectively hydrolyze hemicellulose and further degrade lignin, generating more low-molecular-weight compounds [88]. Song et al. [89] used acidic DESs to pretreat corn stover (CS). At an acid concentration of 4%, lignin removal reached 72.6%, and the cellulose saccharification rate increased to around 80%. Many DESs based on choline chloride (ChCl), including weakly alkaline and acidic DESs, are commonly used for processing various types of biomass, such as wheat straw, corn stover, corn cobs, and rice straw [90]. Chen et al. [91] employed Lewis acid-enhanced DESs at 100 °C, achieving a lignin removal rate of 57.9% and significantly improving enzymatic hydrolysis efficiency from 26.3% to 87.0%. The maximum yield of biohydrogen total solids (TS) was 114.8 mL/g, which is 2.1 times higher than the raw material (37.1 mL/g TS). Based on these findings, DESs can be categorized into four main types (Table 2).

Table 2. General formulas for the classification of DESs [92].

Type	Components	General Formula	
1	Metal salt + organic salt	$Cat^+ X^-$ zMClx	M = Zn, Sn, Fe, Al, Ga, In
2	Metal salt hydrate + organic salt	$Cat^+ X^-$ zMClx. yH_2O	M = Cr, Co, Cu, Ni, Fe
3	HBD + organic salt	$Cat^+ X^-$ zRZ	Z = $CONH_2$, COOH, OH
4	Zinc/aluminum chloride + HBD	MClx + RZ = MCl^+_{x-1}. RZ + MCl^-_{x+1}	M = Al, Zn and Z = $CONH_2$, OH

Note: Cat^+, any ammonium, phosphonium, or sulfonium cation; X, a Lewis base, generally a halide anion; z, the number of y molecules that interact with the anion.

2.4.5. Deep Eutectic Solvent–Alkali Pretreatment

DESs dissolve and partially degrade the cellulose and lignin in biomass, disrupting its crystalline structure and increasing the enzymatic accessibility of cellulose. Following DES pretreatment, alkali treatment further removes lignin and partially dissolves hemicellulose, enhancing the exposed surface area and degradability of cellulose. Song, et al. [89] used alkaline DESs for corn stover (CS) pretreatment, and when the alkali concentration was increased to 4%, lignin removal reached 81.3%, and the enzymatic saccharification rate of cellulose increased to over 85%. DESs composed of hydrogen bond donors (HBD) and acceptors (HBA) have become increasingly significant in biomass pretreatment due to its excellent lignin depolymerization capabilities. Using NaOH, urea, and ethylene glycol to prepare an alkaline DES, the alkaline DES pretreatment significantly improved the methane yield in anaerobic digestion of corn stover (670.3 mL/g VS). A novel fractionation strategy using DES-sodium bicarbonate (DES-SB) for the full component utilization of corn stover (CS) showed that the addition of SB significantly increased lignin removal efficiency (90.03%) compared to pure DES pretreatment (34.64%) and achieved excellent carbohydrate digestibility (glucose yield, 97.47%; xylose yield, 92.93%) [93]. The potential for broader application of DESs in the pretreatment of lignocellulosic organic materials, especially in the energy sector, is promising.

Overall, DESs can selectively dissolve large amounts of lignin while preserving hemicellulose and cellulose as much as possible. Therefore, the DES is expected to play a crucial role in the pretreatment of straw biomass, regarded as a promising and environmentally friendly alternative to traditional solvents, potentially enhancing the conversion efficiency of straw biomass. Several process parameters may influence the efficiency of DES treatment, such as (1) the characteristics of lignocellulosic feedstocks, including their composition, crystallinity, and particle size; (2) the properties of DESs, including the nature of HBA and HBD, as well as their molar ratio; and (3) reaction conditions, including the effects of solid-to-liquid ratio, treatment temperature, and time [1,94]. Therefore, further research into the interaction mechanisms of the DES and alkali, along with optimization of pretreatment parameters, is necessary to improve the overall efficiency and sustainability of biomass conversion.

3. Advantages of Combined Pretreatment

Combining two or more pretreatment methods allows for the synergistic action of chemical agents with physical or biological means, effectively disrupting the structure of lignin and making it easier to remove [52]. For example, acid pretreatment can partially hydrolyze hemicellulose, exposing more lignin surfaces, which can then be more effectively dissolved and removed by subsequent alkali pretreatment [76]. Single-chemical pretreatment methods may produce a significant amount of inhibitors, such as furfural and acetic acid, which can inhibit subsequent enzymatic hydrolysis and fermentation processes [95]. Combined pretreatment can reduce the formation of inhibitors by optimizing the conditions of each method. For instance, a mild acid–alkali pretreatment followed by biological pretreatment can effectively decrease the production of toxic by-products [34].

Combined pretreatment methods can more thoroughly disrupt the complex structure of lignocellulose, particularly the lignin–hemicellulose–cellulose bonds. Physical pretreatments such as steam explosion (SE) or wet oxidation can damage the cell wall structure, enhancing the penetration of chemical reagents and making lignin more accessible for subsequent chemical or biological removal [59]. Combined pretreatment techniques can reduce the amount of chemical reagents needed by using various methods in tandem. For instance, mechanical milling or SE can increase the specific surface area of cellulose, followed by low-concentration acid or alkali pretreatment, which can significantly lower the chemical reagent usage while achieving high lignin removal efficiency [47,49].

Different types of lignocellulosic materials vary greatly in lignin content and structure, and a single-pretreatment method may not be effective for all materials. Combined pretreatment techniques can optimize processing by combining different methods to address the specific characteristics of various raw materials, enhancing lignin removal and increasing the versatility of the pretreatment. The presence of lignin can hinder the enzyme degradation of cellulose and hemicellulose. By effectively removing lignin through combined pretreatment, enzyme efficiency can be significantly improved, leading to higher sugar yields and fermentation efficiency. For example, after acid–alkali combined pretreatment, the residual lignin content is significantly reduced, thereby increasing the rate and efficiency of the enzymatic reaction [74,76] (Figure 4).

Figure 4. Advantages of combined pretreatment.

To explore the economics and environmental impacts of pretreatment, this review provides a qualitative assessment of the energy consumption, cost and environmental impacts of each combined pretreatment method (Table 3). These assessments help to understand the feasibility of different pretreatment methods in practical applications and their impact on sustainable development. In future, when selecting a combined pretreatment technology, not only its technical effectiveness but also the combined energy, economics and environmental impacts need to be assessed in order to achieve a green and efficient biomass pretreatment process.

Table 3. Qualitative assessment of energy consumption, costs and environmental impacts of pretreatment.

Type	Pretreatment Method	Energy Consumption	Cost	Environmental Impact
Physical–Chemical	Steam blast–acid pretreatment	★★★	★★★	★★
	Mechanical crushing–alkali pretreatment	★★	★★	★★
	Moist heat–alkali pretreatment	★★	★★	★★

Table 3. *Cont.*

Type	Pretreatment Method	Energy Consumption	Cost	Environmental Impact
Physical–Biological	Mechanical crushing–biological pretreatment	★★	★★	★
	Steam blasting–biological pretreatment	★★★	★★★	★★
	Moist heat–biological pretreatment	★★	★★	★
Chemical–Biological	Acid–biological pretreatment	★★	★★	★★
	Alkali–Biological Pretreatment	★★	★★	★★
Chemistry–Chemistry	Acid–alkali pretreatment	★★★	★★★	★★★
	Acid-oxidizer pretreatment	★★★	★★★	★★★
	Alkali–oxidant pretreatment	★★★	★★★	★★★
	DES–acid pretreatment	★★	★★★	★★
	DES–alkali pretreatment	★★	★★★	★★

Note: ★ low, ★★ medium, and ★★★ high.

4. Application

Pretreated lignocellulosic materials have a wide range of applications in biorefining. By effectively disrupting the structure of lignocellulose through pretreatment, the accessibility of cellulose and hemicellulose is enhanced, making them more amenable to enzymatic hydrolysis and fermentation, and facilitating their conversion into various high-value products (Figure 5).

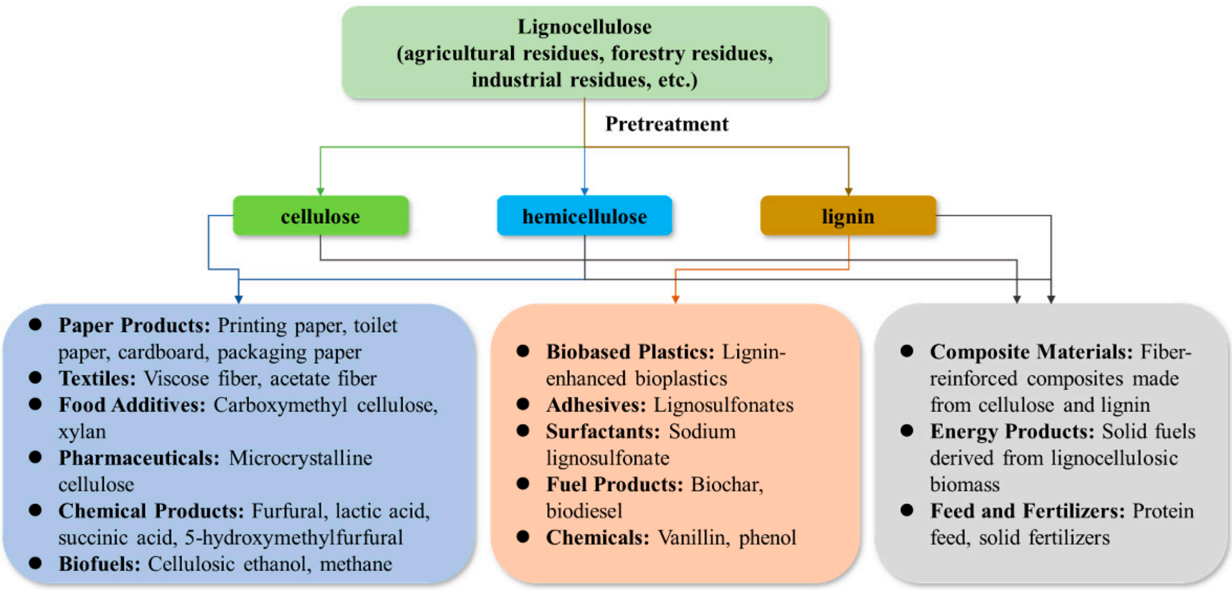

Figure 5. Industrial applications of lignocellulosic components.

4.1. Biofuels

Biofuels are energy products produced from the conversion of biomass resources, such as lignocellulose, vegetable oils, and crop residues, to replace traditional petroleum fuels. Pretreated lignocellulose is initially processed to remove lignin and some hemicellulose through physical, chemical, or biological methods. This pretreatment enhances enzyme accessibility and degradation efficiency, thereby improving the saccharification rate of lignocellulose. After pretreatment, lignocellulose undergoes enzymatic hydrolysis and fermentation to produce fuel ethanol, which is currently the most common application of biofuels. The production of bioethanol generally involves three steps: (1) pretreatment of the raw material, (2) enzymatic hydrolysis of polysaccharides into monosaccharides, and (3) microbial fermentation of monosaccharides into ethanol. Table 4 lists the results of ethanol production from various biomass resources after pretreatment in recent years, Fan et al. [96] used NaOH-catalyzed ethanol pretreatment on poplar to achieve the synergistic maximization of xylose (42.47 g/L) and ethanol (85.74 g/L) yields. This process involved co-producing xylose and ethanol at high solid content, thereby increasing product concentrations while reducing water and energy consumption.

Table 4. Production of fuel ethanol from different types of biomass resources.

Feedstock	Pretreatment	Cellulose Efficiency	Microorganism	Ethanol Concentration	Ethanol Yields	Reference
Cotton stalk	Ultrasound-assisted alkali pretreatment	——	Saccharomyces cerevisiae	——	45.53%	[97]
Corn Stover	Lime-CaO pretreatment	>90%	Saccharomyces cerevisiae CRD61	65.1 g/L	——	[98]
Corn Stover	CaO densification	91.1%	Saccharomyces cerevisiae CRD51	70.6 g/L	——	[99]
Rice straw	KOH/urea	92.02%	Saccharomyces cerevisiae Y2034	37.02 g/L	75.89%	[100]
Reed	Tartaric acid	95.2%	Saccharomyces cerevisiae	10.8 g/L	55.5%	[101]
Reed	LHW-$NH_3 \cdot H_2O/O_2$	97.60%	yeast	71.5 g/L	78.40%	[102]
Oak sawdust	HCl	85.98%	Pichia stipitis KCTC 7222	21 g/L	——	[103]
Rice husk	Alkali-hydrogen peroxide acetic acid (alkali-HPAC)	86.3%	Saccharomyces cerevisiae (KCTC 7906)	——	85.4%	[104]
Sugarcane bagasse (SCB)	PEG 4000 assistance alkaline-catalyzed glycerol	92.1%	Saccharomyces cerevisiae rdna8	56.4 g/L	——	[105]
Poplar	NaOH-catalyzed ethanol	72.84%	S. cerevisiae strain	85.74 g/L		[96]
Poplar	Acetic acid	53.11%	Saccharomyces cerevisiae	30.96 g/L	92.79%	[106]

Lignocellulosic biomass, after pretreatment, can be converted into biogas through anaerobic digestion (AD). Anaerobic digestion of lignocellulosic biomass is a complex process that can be divided into four stages: hydrolysis, fermentation, acidogenesis, and methanogenesis [8]. Pretreatment before AD is a crucial step in the methane production process. Previous reports have shown that although there is a rich source of energy crops, agricultural residues, and other biomasses, pretreatment is necessary to maximize the utilization of raw materials and enhance methane yield [107]. After pretreatment, the recalcitrance of lignocellulosic biomass is reduced, and enzyme accessibility is improved.

Table 5 lists the methane yields of different biomass resources after pretreatment. You et al. [108] reported that the lignin conversion rate increased to 60% and the methane yield from anaerobic digestion rose to over 500 mL/g TS using ultrasonic–CaO–NaOH pretreatment. This indicates that physical–chemical combined pretreatment results in higher lignin conversion rates, thereby improving the cost-effectiveness of biogas production.

Compared to traditional fossil fuels, bioethanol and biogas offer lower carbon emissions and greater environmental sustainability. Biofuels are renewable energy sources that can reduce dependence on finite oil resources, thereby mitigating risks related to energy security and environmental issues. In addition to being used in transportation, biofuels can also be employed in electricity generation, heating, and other areas, contributing to energy diversification. By employing combined pretreatment of lignocellulosic biomass, the efficiency and economic viability of biofuel production can be enhanced, promoting the sustainable development of biomass energy, advancing environmental protection, and reducing greenhouse gas emissions. This holds significant socio-economic and environmental importance.

Table 5. Methane production from various biomass resources.

Feedstock	Pretreatment	Bio-Methane Production	Reference
wheat straw	Mechanical grinding–hydrothermal	376 mL/g VS	[109]
Corn straw	NaOH pretreatment with CaO additive and ultrasound	500 mL/g-TS	[108]
Corn straw	Grinding–urea	250.03 mL/g VS	[7]
Cyperus papyrus 'Nanus'	Ball milling–hydrothermal	180.57 mL/g VS	[110]
Corn straw	Grinding–alkaline densification	224.30 mL/g VS	[111]
Wheat straw	Grinding–urea	305.5 mL/g VS	[112]
Rice straw	Mechanical grinding–nanobubble water	336.7 NmL/g VS	[113]

4.2. Bio-Based Chemicals

Bio-based chemicals are produced using biomass resources such as lignocellulose, plant oils, and biomass residues, offering higher renewability and environmental friendliness compared to traditional petroleum-based chemicals.

After pretreatment, lignocellulose undergoes enzymatic or acid–alkaline treatment to release sugars containing organic acids. These sugars can be fermented by microorganisms to produce various organic acids, such as lactic acid and succinic acid. For instance, pretreatment of corn straw with solid acid under optimal conditions (digestion temperature of 120 °C, digestion time of 80 min, and solid acid concentration of 1.5%) achieved a glucose conversion rate of 71.06%. Using *Lactobacillus delbrueckii* as the starter strain for D-lactic acid production, a yield of 18 g/L and optical purity of 99% were obtained [114]. Zhang et al. [115] developed a simultaneous saccharification and fermentation (SSF) process, which involved pretreating mechanically shredded corn straw with 15% NaOH. The process used 30 FPU (filter paper units)/g cellulase and 20 g/L corn steep powder in a 5-L bioreactor to produce lactic acid (LA). The resulting lactic acid concentration, yield, and productivity were 104.11 g/L, 0.69 g/g, and 1.24 g/L/h, respectively. Using straw, a renewable biomass resource, for lactic acid production through pretreatment and fermentation processes aligns with sustainable development goals by reducing dependence on non-renewable resources and improving resource utilization efficiency. Dąbkowska et al. [116] employed an organic solvent method (80% glycerol and 1.25% H_2SO_4) to successfully pretreat ground Miscanthus, achieving high sugar recovery rates (glucan > 98%, xylan > 91%) and biomass delignification rates (60%). After fermentation of the hydrolysate with *Actinobacillus succinogenes* 130Z, the succinic acid yield was 75–82%. The biotechnological conversion of inexpensive lignocellulose into high-value organic acids, such as lactic acid for biodegradable plastics (PLA) and succinic acid as a platform compound for various chemical syntheses, offers significant economic benefits.

Lignocellulose, after pretreatment, can be converted into platform chemicals such as furfural and hydroxymethylfurfural (HMF). Furfural can be used to produce solvents, resins, and fuel additives, and can be further transformed into chemicals like furan-2,5-dicarboxylic acid (FDCA) and adipic acid. Li et al. [117] developed a two-step pretreatment process for separating and producing various products (furfural, ethanol, and lignin) from corn straw (CS). In the first step, H_2SO_4 pretreatment was used to remove hemicellulose. The resulting hemicellulose-containing wash liquor was used to produce furfural, achieving up to 4.0 g/100 g CS. The subsequent NaOH pretreatment removed 90.8% of the lignin. Avci et al. [118] achieved a furfural yield of 10.8 ± 0.3 g/100 g straw, which corresponds to 61.6% of the theoretical yield, under optimal conditions (200 °C, 0.75% (v/v) acid concentration) with a pretreatment time of 20–25 min for 5% (w/w) corn straw. Furfural is a key intermediate for producing various chemical products, including furfuryl alcohol, tetrahydrofuran (THF), and 2-methylfuran. Converting agricultural waste into high-value furfural enhances the economic value of biomass resources and promotes the added-value utilization of agricultural waste.

Hydroxymethylfurfural (HMF) is an important bio-based platform chemical that can be further converted into various high-value chemicals such as 2,5-furandicarboxylic acid (FDCA), 2,5-dimethylfuran (DMF), and adipic acid. Li et al. [119] used a combined ultrasound-ionic liquid-ion exchange resin catalyst pretreatment for sugarcane bagasse. Under the conditions of the solvent [Bmim]OAc, the catalyst D001-cc, a treatment time of 25 min, and a temperature of 140 °C, the yield of 5-HMF reached 65.72%. Jasmine et al. [120] pretreated rice straw with microwave-assisted sodium hydroxide. After enzymatic hydrolysis, the maximum reducing sugar yield from rice straw (TRS) was 350 mg/g. Using titanium magnetic silica nanoparticles as a catalyst for microwave-assisted syrup conversion, under microwave irradiation at 120 °C for 30 min, the yield of 5-HMF was 41.1%. The production of HMF from lignocellulose is significant as it facilitates the efficient utilization of renewable resources, promoting sustainable development. Research into pretreatment methods for generating platform chemicals from lignocellulose not only improves the utilization of biomass resources but also advances green chemistry and renewable energy, reduces dependence on non-renewable resources, and supports the development of a circular economy.

4.3. Feed and Fertilizers

Lignocellulose, when subjected to appropriate pretreatment (such as biological, physical, or chemical treatments), can effectively degrade lignin and some hemicellulose, thus enhancing the availability of cellulose. Pretreated lignocellulose can be hydrolyzed to produce sugar-rich products, which can serve as additives in animal feed. Xylooligosaccharides (XOS), a type of oligosaccharide composed of 2–10 xylose units, are widely used in the food and feed industries. By hydrolyzing mechanically ground poplar wood with NaHSO4, under optimal conditions of 170 °C for 60 min, an XOS yield of 42.7% was achieved. The hydrolysate rich in XOS can be used directly as a feed additive without the need for NaHSO4 separation [121]. Alkaline hydrothermal pretreatment of sawdust resulted in a purity of 77% for low-xylan content, reaching 62.5% [122]. XOS are high-quality prebiotics that selectively promote the growth of beneficial gut bacteria (such as Bifidobacteria and Lactobacilli), inhibit harmful bacteria, thereby improving intestinal health and reducing disease incidence. They optimize the digestive system, enhance enzyme activity, and increase the absorption of nutrients.

Pretreated lignocellulose residues can be converted into organic fertilizers through biodegradation and fermentation, and have broad applications in crop cultivation, horticulture, and greenhouse vegetable production [123]. These organic fertilizers are rich in carbohydrates, nitrogen, phosphorus, potassium, and other nutrients, which can enhance soil fertility and increase crop yield [124]. For example, the hydrothermal pretreatment and anaerobic digestion of sargassum promote the degradation of organic particles in sargassum, resulting in a maximum soluble chemical oxygen demand (COD) of 27,250 mg/L, which is 237% higher than untreated biomass. This indicates that most organic matter is consumed, and the digested residue is pathogen-free, nutrient-rich, and has potential as a biofertilizer [125]. Xie, et al. [7] used solid urea to pretreat ground corn straw for 2 weeks, finding that the anaerobic digestion residue had a 9.62% higher yield compared to the control, with heavy metal content remaining within safe limits, making it suitable for organic fertilizer production. Utilizing pretreatment products of lignocellulose for feed and fertilizer production effectively uses biomass resources, reduces production costs in agriculture and livestock, and contributes to environmental protection by decreasing fossil energy use and greenhouse gas emissions. Producing high-quality feed and fertilizers improves livestock efficiency and crop yields, contributing to sustainable agricultural development.

In summary, the technology of combined pretreatment of lignocellulose has significant application prospects and socio-economic benefits in feed and fertilizer production, playing an important role in advancing sustainable development in agriculture and livestock.

4.4. Integration of Biorefinery Processes

In biorefinery processes, pretreated lignocellulose can simultaneously produce multiple products through process integration, enhancing resource utilization efficiency and achieving waste minimization and value maximization. For example, hydrothermal treatment of rapeseed straw efficiently produces reducing sugars and XOS [126]. The hydrothermal treatment using water as a solvent and catalyst disrupts the dense structure of rapeseed straw, increasing its enzymatic hydrolysis efficiency from 24.6% to 92.0%. After treatment at 200 °C for 60 min, XOS are obtained at 3.3 g/L. Chang et al. [127] used extrusion combined with biological pretreatment on Glycyrrhiza uralensis residue (GUR), achieving an enzymatic hydrolysis rate of 81.06%. From 100 g of GUR, 1.49 g of flavonoids, 294.36 U of cellulase, and 14.13 g of ethanol can be produced. The key aspects of biorefinery process integration are:

(1) Multi-Technology Integration: Combining physical, chemical, and biological technologies to select appropriate treatment and conversion paths based on the characteristics of the biomass resources.
(2) Efficient Energy Utilization: Optimizing production processes to enhance energy utilization efficiency and product selectivity, reducing production costs and environmental impact.
(3) Sustainable Development: Promoting the development of the bio-economy, reducing dependence on finite resources, lowering greenhouse gas emissions, and advancing the establishment of a circular economy model.

The success of biorefinery process integration depends on the ability to innovate technologies and optimize processes, which requires interdisciplinary team collaboration, combining knowledge and technologies from chemical engineering, bioengineering, environmental science, and other fields to achieve comprehensive utilization and industrial application of biomass resources.

5. Combined Pretreatment Future Development Strategy and Outlook

With the continued advancement and innovation in pretreatment technologies, more efficient and environmentally friendly methods are expected to emerge, improving resource utilization efficiency and product selectivity.

This review identified and summarized the key advantages and bottlenecks of combined pretreatment technologies in lignocellulosic biomass degradation, in particular the potential of synergistic action of multiple pretreatment methods in improving cellulose accessibility, conversion efficiency and environmental friendliness. Compared with single-pretreatment methods, combined pretreatment methods can significantly improve reaction conditions, reduce chemical usage, and optimize subsequent enzymatic and fermentation processes, which provides new technological pathways for industrial applications. The application of combined pretreatment technologies will promote wider utilization of biomass resources and expand market opportunities in areas such as biofuels, bio-based chemicals and biomaterials. Compared with other literature reviews, this review is unique in that it comprehensively and systematically analyzes the effects of different combined pretreatment methods, especially the advantages and disadvantages of different pretreatment technologies, from the perspective of interdisciplinary intersection of physics, chemistry, and biology.

In the future, interdisciplinary co-operation will continue to promote the innovation and application of combined pretreatment technologies, making full use of the strengths of chemical engineering, bioengineering, materials science and other disciplines to promote technological advances and industrial applications. Particularly in focusing on the application prospects and feasibility of combinatorial technologies, this review comprehensively guides the efficient utilization of biomass resources and lays a theoretical foundation for future technology optimization and sustainable development strategies. Combined pretreatment technology shows great technological advantages and market potential in the utilization of biomass resources.

6. Conclusions

Lignocellulosic biomass is one of the most abundant renewable resources on the planet, but its complex structure hinders its efficient conversion to high-value products. The aim of this study was to analyze and summarize recent advances in combined pretreatment technologies that integrate physical, chemical, and biological methods to improve the efficiency of subsequent conversion. The results show that combined pretreatment methods can significantly improve biomass conversion efficiency and yield. Notable advances include the selective breakdown of lignocellulosic components into valuable sugars and platform chemicals while generating specific products such as organic acids and furfural. However, many of the combined pretreatment technologies are still in the pilot stage and scalability challenges remain. In addition, high operating costs and limited long-term environmental impact data are areas for further exploration. In summary, combined pretreatment technologies offer great potential for advancing biomass utilization and could make a meaningful contribution to achieving sustainable energy and environmental goals.

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