Review Paper

Pelletization can unlock the unrealized potential of lignocellulose as a resilient feedstock for biomanufacturing: enzymatic saccharification of biomass pellets

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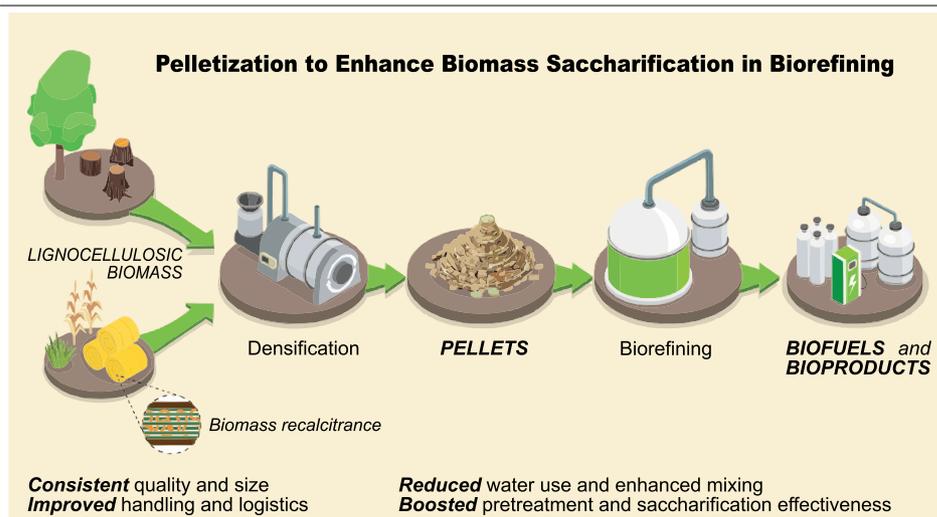
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HIGHLIGHTS

➢Pelleting serves as a bridge between upstream logistics and downstream bioconversion.
➢Pellet attributes shaped by process variables influence the yield of sugar release.
➢Saccharification of pellets is driven by pellet traits and conversion settings.
➢Energy use, costs, and environmental impacts are evaluated for pellet bioconversion.
➢Advances in pelleting technologies optimize biofuel production and feedstock utilization.

GRAPHICAL ABSTRACT

Pelletization to Enhance Biomass Saccharification in Biorefining

LIGNOCELLULOSIC BIOMASS → Densification → **PELLETS** → Biorefining → **BIOFUELS** and **BIOPRODUCTS**

Biomass recalcitrance

Consistent quality and size
Improved handling and logistics
Reduced water use and enhanced mixing
Boosted pretreatment and saccharification effectiveness

ARTICLE INFO

ABSTRACT

Lignocellulose, as a plentiful and renewable carbonaceous resource, presents an alluring alternative to fossil fuels for sustaining industries in the pursuit of a resilient bio-based economy. Sugars derived from lignocellulosic biomass play a central role as versatile platform intermediates for feeding microorganisms or as starting chemicals for manufacturing value-added fuels, chemicals, and materials. However, commercialization faces challenges due to the complexity and high costs associated with feedstock logistics and conversion processes. Pelleting offers a potential solution by addressing logistical issues while providing additional benefits for downstream conversion that may outweigh the extra costs associated with pelleting. To fully unlock the economic and sustainable potential of lignocellulosic biomass in biorefineries, recent advances in pelleting technologies and their impacts on downstream pretreatments and enzyme-mediated conversion are critically reviewed. Pelleting has been shown to improve enzymatic digestibility yields by 5–20%. The process variables, product attributes, and their influences on bioconversion are discussed. More significantly, a thorough discussion of the effect of pelleting on various pretreatments, concerning diverse feedstocks, as well as their interplay, is provided to inform the design of future pelleting and pretreatment processes. Finally, practical considerations, including energy consumption, costs, and environmental impacts, are discussed, alongside an exploration of cutting-edge technologies and strategies in this field.

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Contents

Abbreviations

AFEX	Ammonia Fiber Expansion
AI	Artificial Intelligence
DES	Deep Eutectic Solvent
DLC	Densifying Lignocellulosic biomass with Chemicals
DLCA	Densifying Lignocellulosic biomass with Chemicals under Autoclave
GHG	Greenhouse Gas
IL	Ionic Liquid
LCA	Life Cycle Assessment
MESP	Minimum Ethanol Selling Price
PEG	Polyethylene Glycol
SAA	Soaking in Aqueous Ammonia
TEA	Techno-economic Analysis
XU	Xylanase Unit

1. Introduction

Rising demand for advanced fuels, chemicals, and materials makes renewable feedstocks central to resilient supply chains and bio-based industries (Liu et al., 2021; Mujtaba et al., 2023). Lignocellulosic biomass, the most plentiful nonfood carbon resource from photosynthesis on Earth, is often discarded as waste yet remains a crucial complement to fossil feedstocks in a multi-pronged strategy toward energy security (Lynd et al., 2022; Chen et al., 2024). Biorefining converts lignocellulose into high-value fuels and products through either thermochemical processes, which produce syngas or bio-oil (Huber et al., 2006), or through biochemical processes, which release fermentable sugars (Ding et al., 2024). The latter holds considerable promise because water-soluble sugars are versatile platform intermediates for producing a broad spectrum of commercially relevant molecules that are competitive with petrochemicals (Ragauskas et al., 2006; Zhou et al., 2021). Examples include biomass-derived carbohydrates serving as a carbon source to feed microorganisms for biomanufacturing renewable fuels, chemicals, and polymers (Scown, 2022; He et al., 2023), or as a starting material for generating diverse commodity chemicals and/or precursors of value-added molecules such as furans, sugar alcohols, and organic acids (Bozell and Petersen, 2010; Chen et al., 2018; Zhang et al., 2021).

Sugars are typically extracted from lignocellulosic biomass through hydrolysis, also known as saccharification, to break down its complex polysaccharides, such as cellulose and hemicellulose, into their constituent monosaccharides using either acids or enzymes (Zhou et al., 2021; Lyu et al., 2022; Zhu and Pan, 2022). While acid hydrolysis is a relatively fast and straightforward means for obtaining biomass-derived sugars, enzymatic saccharification is more advantageous for selectively producing higher-quality fermentable sugars while circumventing the need for harsh chemicals and conditions, costly purification steps to remove degradation products, and limiting potential environmental impacts (Ladisch et al., 1983; Zhai et al., 2022). Additionally, advances in biotechnology are being used to develop more productive and cost-competitive enzymes for biomass transformation, which could further enhance the efficiency and sustainability of enzymatic saccharification (Payne et al., 2015). Nevertheless, the commercialization of lignocellulose's potential for biomanufacturing *via* a sugar platform has not yet taken off, mainly due to the significant costs associated with biomass feedstock and conversion processes.

Conventional pretreatment methods such as dilute acid, steam explosion, liquid hot water, lime, and ammonia have been widely applied to disrupt the lignocellulosic structure and enhance enzymatic accessibility (Mosier et al., 2005; Ding et al., 2024). Emerging methods are being developed to simplify processing, reduce costs, and improve enzymatic saccharification performance, such as deacetylation and mechanical refining (Chen et al., 2016), biocompatible ionic liquids (Xu et al., 2016; Krishnamoorthy et al., 2025), deep eutectic solvents (Verdía Barbará et al., 2025), and other advanced solvents (Baral et al., 2025; Chen et al., 2025). Instead of relying on chemicals, solvents, or high temperatures, mechanical pretreatments such as extrusion (Duque et al., 2017), ball milling (Chen et al., 2022b; He et al., 2023), and pelletization (Chen et al., 2023a) modify structural and physicochemical properties through physical forces to improve digestibility.

Beyond conversion, the deployment of lignocellulosic biorefineries is critically limited by upstream logistics. Unlike petroleum, which is naturally concentrated in specific locations and has higher energy and carbon densities, lignocellulosic biomass is widely dispersed, bulky, and of low energy density, which complicates collection, storage, and long-distance transport (Tumuluru et al., 2011; Li et al., 2020). Additionally, the non-uniform size and shape of the biomass pose challenges for storage and handling, particularly in large-scale operations. The poor flow properties and low bulk density of loose or baled biomass necessitate biorefineries to invest in supplementary conveyor systems and storage facilities, which incur additional costs, including labor expenses, thereby negatively affecting the economics of handling and storing the feedstock (Theerarattananoon et al., 2012). In addition to raising the cost of raw material supply, these characteristics prevent biomass from functioning as a true commodity feedstock, forcing biorefineries to depend on locally

sourced agricultural residues.

Pelleting offers a unique opportunity to address both dimensions, biomass recalcitrance and logistics (Fig. 1). As a densification strategy, pelleting increases bulk density up to tenfold (Theerarattananoon et al., 2012), improves size uniformity and grain-like flowability, and enhances durability, thereby reducing transportation and storage costs and enabling the use of existing grain-handling infrastructure (Theerarattananoon et al., 2011; Guragain et al., 2013). At the same time, high shearing forces and frictional heating during pelleting act as a mild thermochemical pretreatment, reducing the natural recalcitrance of lignocellulose to enzymatic hydrolysis—a major obstacle in utilizing lignocellulose (Chen et al., 2023a; Gong et al., 2023). Nonetheless, some studies have observed adverse effects of pelleting on bioconversion (Theerarattananoon et al., 2012; Gong et al., 2021b), which can be attributed to variations in feedstock characteristics, pretreatment, pelleting processes, and their combinations. Therefore, understanding how to economically and sustainably produce pellets that are favorable for both logistics and bioconversion is crucial in unlocking the unrealized potential of pelleting for efficient and profitable lignocellulosic sugar production, to decarbonize the chemical industry.

This review aims to provide a systematic overview of research on utilizing biomass pellets for sugar production, offering a comprehensive understanding of the fundamentals and technological gaps in pelleting biomass for future biorefinery applications (Table 1). It covers various aspects of the pelleting process, including techniques, variables, and attributes that affect the practical aspects of generating pellets from loose

biomass, as well as aspects of the process that positively and negatively impact downstream saccharification and fermentation of the resulting pellets into fuels and biochemicals. Considering the downstream effects of pelleting is essential for advancing pelleting and conversion technologies. Additionally, we compare diverse pelleting scenarios, discuss practical considerations such as energy consumption, and evaluate economic and environmental perspectives while exploring innovative technologies and strategies.

2. Lignocellulosic biomass and sugars

Lignocellulosic biomass, sometimes referred to as cellulosic biomass, plant biomass, or lignocellulose, is the fibrous, inedible, and usually discarded part of plants that are mainly grown for food, feed, and fiber production. It provides plants with rigidity and structure and comprises three primary carbon-based polymers: cellulose, hemicellulose, and lignin, collectively known as lignocellulose, which stores the solar energy captured through photosynthesis in the form of plant cell walls (Rubin, 2008). Cellulose, the most abundant biopolymer on Earth, is a linear homopolysaccharide composed of glucose molecules linked by glycosidic bonds. Hemicelluloses, on the other hand, are branched and heterogeneous carbohydrate polymers of glucose or xylose, substituted with various 5- and 6-carbon monosaccharides, and may also contain side chains with ferulate esters or acetyl groups (Scheller and Ulvskov, 2010). Aligned bundles of highly crystalline cellulose fibrils surrounded by a disordered matrix of
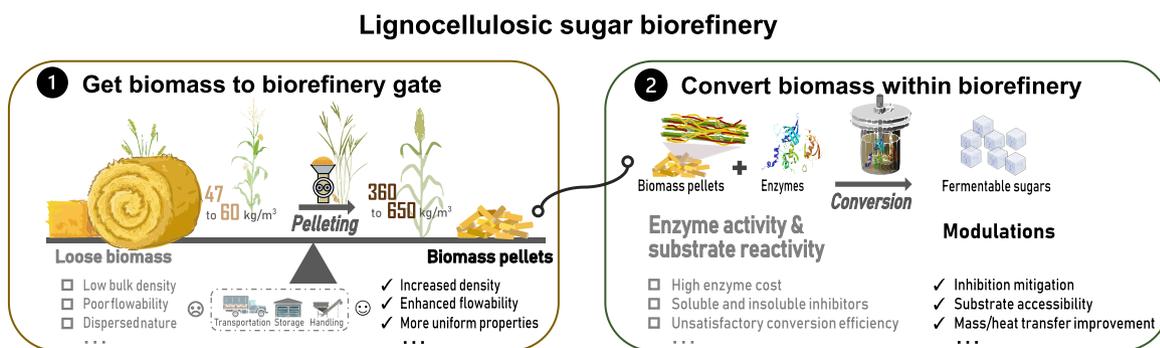
Lignocellulosic sugar biorefinery

Fig. 1. Pelleting connects upstream logistics with downstream bioconversion in a lignocellulosic sugar biorefinery. Data sourced from Theerarattananoon et al. (2012); part of the images from BioRender.

Table 1.
Comparison of key features across recent review studies on pelletization of lignocellulosic biomass for biochemical conversion.

Feedstocks	Pelletization/Densification Focus	Biochemical Conversion Coverage	Key Contributions	Ref.
Lignocellulosic residues (wheat straw, corn stover, bagasse, poplar, grasses)	Reviews densification methods; particle size, durability, and logistics cost	Detailed on routes to sugars; pelletization impact on hydrolysis & fermentation; cites lab studies	Provides structured evaluation of how particle size reduction + pelletization influence sugar yields; identifies merits/limitations	Tumuluru, (2020a)
Softwood chips and pellets	Focus on pellet/chip feed forms; densification impact on lignin relocation	Investigates lignin relocation/modification and enzymatic hydrolysis; explains softwood recalcitrance (lignin content/structure)	Demonstrates that Chemithermomechanical Pulping (CTMP)-based pretreatment modifies/relocates lignin, enhancing hemicellulose recovery and cellulose hydrolysis of chips and pellets	Takada et al. (2020)
Lignocellulosic residues (woody + herbaceous)	Pellets & briquettes; process variables; emphasizes logistics efficiency	Reviews pretreatments applied to pellets; summarizes glucose/ethanol yields	Consolidates numerous experimental results showing pelleting + pretreatment can enhance hydrolysis rates and reduce enzyme loadings	Paul et al. (2023)
Lignocellulosic biomass	Densification processes; parameters & quality; supply chain	Addresses enzymatic hydrolysis post-pelleting; highlights neutral/positive impacts when combined with pretreatments	First critical synthesis of densification–pretreatment integration in biorefineries; different pelleting strategies; techno-economic analysis	Takada et al. (2020); Gong et al. (2023)
Agricultural residues, energy crops, woody biomass	Systematically compares pelletization variables and feedstock traits; analyzes diverse pelleting strategies; emphasizes advanced pelleting for reduced cost/energy	Direct comparison of saccharification for pelleted vs. non-pelleted biomass across feedstocks, pretreatment types, and conditions, with and without pretreatment; includes novel pelletization-based pretreatment	(i) Frames pelletization as both a logistics solution and a pretreatment enhancer; (ii) quantitatively compares sugar yields from pelleted and unpelleted biomass, both with and without pretreatment, in overview figures; and (iii) unifies diverse studies on technologies, variables, attributes, yields, and techno-economic/environmental impacts into a coherent pelleting-based biorefinery framework	**This Study**

hemicellulose and lignin, a three-dimensional polymer of phenylpropanoid units, make the sugars difficult to access by hydrolytic enzymes and resistant to deconstruction (Petridis and Smith, 2018).

Efficient, cost-competitive, and environmentally friendly extraction platforms for sugars from lignocellulosic biomass remain a central challenge to their utilization. Generally, the polysaccharides (cellulose and hemicellulose) in lignocellulose are hydrolyzed by chemical (acid) or enzyme catalysts to obtain the soluble carbohydrates (fermentable sugars) (Zhu and Pan, 2022). Acid-catalyzed hydrolysis has a long history as an efficient and facile approach for releasing sugars from lignocellulose (Binder and Raines, 2010). Concentrated acids deliver high sugar yields under mild temperatures but face challenges in separation and recycling (Wolfaardt et al., 2021). In contrast, dilute acids require high temperatures that cause sugar degradation, necessitating optimization through kinetic control, reactor design, and process integration (Zhou et al., 2021). Solid acids offer easier recovery and reduced corrosion but remain limited by low activity, poor substrate affinity, and insufficient durability (Zeng and Pan, 2022). It is noteworthy that during the acid hydrolysis of lignocellulose to release monosaccharides, the cleavage of lignin linkages is unavoidable, often leading to its simultaneous depolymerization and repolymerization, which in turn results in the formation of undesirable lignin-derived phenolics and highly condensed lignin structures (Zhu and Pan, 2022).

Alternatively, enzymatic hydrolysis offers remarkable selectivity in liberating sugars from lignocellulose, while having minimal influence on lignin and generating negligible downstream byproducts (Jørgensen et al., 2007; Payne et al., 2015). Acting in a heterogeneous system, cellulases, hemicellulases, and accessory enzymes cooperatively depolymerize cellulose and hemicellulose, yielding fermentable sugars of high purity (Payne et al., 2015; Zhai et al., 2022). Compared to chemical saccharification, enzyme-mediated hydrolysis operates under mild, non-corrosive conditions, thereby reducing utility demands and equipment costs. Together with the use of biodegradable enzymes derived from renewable sources (Payne et al., 2015), enzymatic hydrolysis reduces the formation of harmful waste and minimizes energy requirements for waste disposal, thereby reducing associated GHG emissions. Advances in biotechnology continue to improve enzyme efficiency and affordability (Guo et al., 2022). Yet, high enzyme costs and the need for effective pretreatment remain major barriers, as pretreatment is essential to overcome lignocellulose's inherent recalcitrance (Mosier et al., 2005).

3. Pelleting of lignocellulosic biomass

The successful utilization of lignocellulose for fermentable sugar production requires not only an effective conversion strategy to extract sugars from biomass but also a reliable feedstock logistics platform that ensures a sustainable and cost-effective supply throughout the year (Morais et al., 2022). However, the low bulk density and poor flow characteristics of conventional chopped or baled biomass make it challenging to store, transport, and process, limiting its availability for conversion to products at scale (Mamun et al., 2020). An appealing solution to this challenge involves densifying loose biomass into uniform, commodity-type feedstocks while increasing unit density up to tenfold through processes such as pelleting, briquetting, extrusion, or agglomeration (Table 2). These densification approaches were originally designed for solid fuel generation and have been adapted from other biofeedstock processing industries, including feed, food, and pharmaceuticals. Among them, pelleting and briquetting are widely employed to manufacture densified biomass for use as combustible fuel (Tumuluru et al., 2011; Li et al., 2014). A pellet mill is advantageous due to its high efficiency and effectiveness in compaction, ease of machine maintenance, and resulting pellets that exhibit better handling features compared to briquettes (Table 2) (Dien et al., 2018). These factors have contributed to the rapid increase in global production and consumption of pelleted biomass as a tradable commodity (Takada et al., 2020). Moreover, pelleted biomass shares flowability characteristics with cereal grains, allowing it to be transported, handled, and stored using existing equipment designed for use with grains throughout the entire biomass supply chain, including field operations, road transportation, and within biorefinery facilities (Theerarattananoon et al., 2011). The grain-like features of pelleted biomass prompt existing grain-processing biorefineries to transition from grains to lignocellulosic materials as the primary feedstock, due to the potential to utilize available grain storage and processing infrastructure for biomass pellets, thereby minimizing the need for additional capital investment (Guragain et al., 2013).

3.1. Pelleting

Pelleting, a process widely employed in the feed, fuel, and pharmaceutical industries, utilizes mechanical or thermal processing means to agglomerate small particles into larger ones. It converts loose ingredients into dense, free-flowing, and durable pellets using specialized pellet mills. These pellet mills typically comprise a perforated hard steel die with two or more rollers and can be categorized into two types: flat die and ring die (Fig. 2) (Tumuluru, 2020b). The operating principle is the same for both types, where the die and rollers rotate to compress and extrude the feedstock through the die holes. The free flow of the feed is impeded by the friction between the die and the fed-in feedstock particles, resulting in particle compression within the die and the formation of uniform cylindrical shapes with 4–12 mm or even larger in diameter and varying lengths (generally less

Table 2.
Comparison of densification technologies for lignocellulosic biomass (Tumuluru, 2020b).

Aspect	Pellet Mill	Briquette Press	Extruder	Roller Press	Agglomerator
Pelleting moisture range (%, w.b.)	10–38	10–18	10–40	10–15	40–50
Particle size	Medium to larger (4.76–12.07 mm)	Medium to larger (4.76–19.05 mm)	Smaller (4.76–6.35 mm)	Medium to larger (4.76–12.07 mm)	Smaller (135–397 μm)
Wear of contact parts	High	Low	High	High	Low
Output from the machine	Continuous	In strokes	Continuous	Continuous	Continuous
Specific energy consumption (kWh/t)	16.4–74.5	37.4–77	36.8–150	29.91–83.1	No information
Throughputs (tonne/h)	5	2.5	5	5–10	5
Density of briquette (g/cm³)	0.7–0.8	1–1.2	1–1.4	0.6–0.7	0.4–0.5
Maintenance	Low	High	Low	Low	Low
Combustion performance of briquettes	Very good	Moderate	Very good	Moderate	No information
Carbonization of the feedstock during densification	Not possible	Not possible	Makes good charcoal	Not possible	Not possible
Suitability in gasifiers	Suitable	Suitable	Suitable	Suitable	Suitable
Suitability for cofiring	Suitable	Suitable	Suitable	Suitable	Suitable
Suitability for biochemical conversion	Suitable	Suitable	Not suitable	Suitable	No information
Homogeneity of densified biomass	Homogeneous	Not homogeneous	Homogenous	Not homogeneous	Homogeneous

than five-fold diameter) (García-Maraver et al., 2011), and unit density of 1125–1190 kg/m³ (Tumuluru et al., 2011). However, there are differences in feeding manners, production capacity, energy efficiency and many other aspects between flat die and ring die pellet mills (Tumuluru, 2020b), which are critical to consider when selecting the appropriate pellet mill for a given lignocellulosic feedstock.
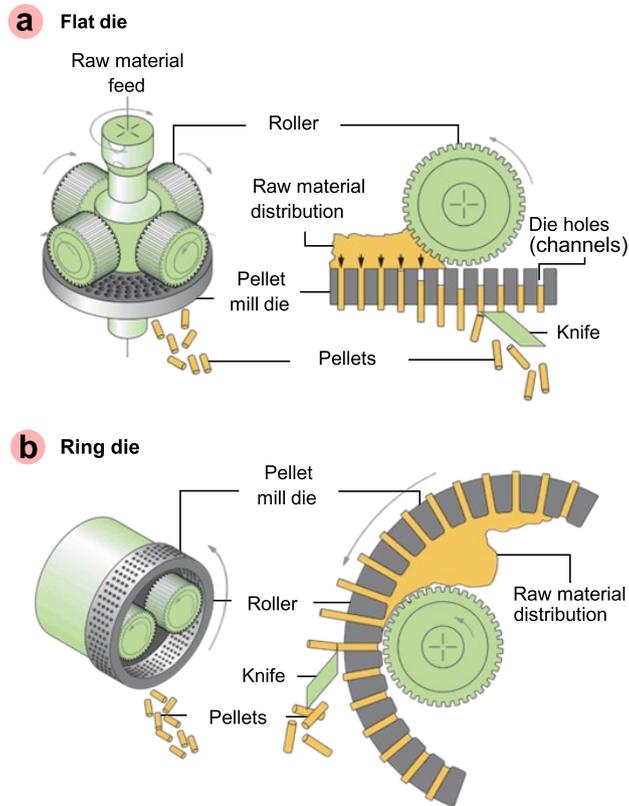

Fig. 2. Designs and operating principles of **(a)** flat die pellet mill (adapted from Tumuluru (2020b), Figure 2.9, p. 31; Springer Nature) and **(b)** ring die pellet mill (adapted from Tumuluru (2020b), Figure 2.10, p. 32; Springer Nature).

3.2. Pelleting variables and impacts

The characteristics of biomass pellets, such as density, durability, and quality, are influenced by both process parameters and feedstock features (**Fig. 3**) (Tumuluru et al., 2011). Significant process variables include temperature, pressure, as well as die geometry and speed. Heat and pressure during the pelleting of lignocellulose could cause the relocation of lignin, which acts as a natural binder for pellet formation (Kaliyan and Morey, 2010) and affects the bioconversion efficiency (Gong et al., 2021b). Die geometry, i.e., size and shape, influences process attributes (e.g., pelleting capacity and energy demand) and product properties (e.g., density and durability). The die's length-to-diameter (L/D) ratio, or compression ratio, representing the proportion of die thickness to die hole (channel) diameter, is an essential indicator of pelleting compression extent. Thicker dies increase pressure, while wider holes decrease pressure (Holm et al., 2006). An increase in L/D ratio results in higher pellet density, strength, and durability, as it leads to greater shear force (Kaliyan and Vance Morey, 2009; Theerarattananoon et al., 2011; Tumuluru, 2023) and influences the pretreatment of pelleted biomass (Theerarattananoon et al., 2012). The speed of the pellet die has a notable impact on pelleting capacity and energy consumption, as well as product characteristics, by altering the material's retention time in the die. Higher speeds increase throughput but also raise energy use due to greater motor power and friction. A study on high-

moisture corn stover pelleting found that optimal speeds (50–60 Hz) maximize density and durability through enhanced compression and lignin plasticization, as determined by response surface modeling. This modeling also revealed interactions with feedstock moisture and preheating (Tumuluru, 2014). Suboptimal speeds (<50 Hz) limit binding and heat transfer, reducing pellet density and durability while increasing energy use per kg. Excessive speeds (greater than 60 Hz) pose a risk of uneven feeding, pellet cracking, and energy spikes. Aligning die speed with moisture and temperature optimizes pellet quality and energy efficiency.

Feedstock properties, including moisture content, particle dimensions, and chemical composition, are also crucial for the pelleting process and pellet quality (Tumuluru et al., 2011; Tumuluru and Yancey, 2018). The moisture content is known to affect biomass densification through various mechanisms, including lowering the glass transition temperature, facilitating solid-bridge formation, and enhancing particle contact area *via* van der Waals forces (Tumuluru et al., 2011; Tumuluru, 2014). However, its effect varies with other feedstock features and process parameters. While an optimum moisture level of 8–12% was suggested for pelleting cellulosic materials (Sokhansanj et al., 2005), a high moisture range of 28–38% has been shown to improve the physical properties of corn stover pellets (Tumuluru, 2014; Tumuluru, 2023). Besides impacting the pellet's physical properties, moisture within biomass may generate steam under elevated pressure, which is likely to decompose hemicellulose and lignin into smaller oligomers of carbohydrates, phenolics, and other derivatives (Li et al., 2014; Chen et al., 2023b). Additionally, a smaller particle size of biomass feedstock is helpful for the pelleting process (Tumuluru, 2023), which is also favored by a broader particle size distribution (Kaliyan and Vance Morey, 2009). Furthermore, the quality of pellets is closely linked to the chemical composition of the feedstock, which contains natural binders such as lignin and protein (Kaliyan and Morey, 2010). These feedstock characteristics and process parameters are the interacting factors determining pellet attributes (Tumuluru, 2014).

As these natural binders are critical to increased binding and bridging among the lignocellulosic molecules found in biomass, it is worth briefly considering the relevance of non-naturally occurring additives, which can have similar impacts on pellet formation and quality. Additives, such as pyrolysis oils, have been shown to reduce friction during the densification process, thereby significantly reducing specific energy requirements (Sarker et al., 2021). In addition to lowering pelleting costs, numerous studies have demonstrated that various additives affect pellet characteristics, including density, durability, higher heating value, and ash content (Tumuluru et al., 2011; Tumuluru, 2020b). For example, increasing the starch content enhances both density and durability due to starch's ability to bind biomass particles through gelatinization (Tumuluru et al., 2016). Lignin, a commonly used binder, contributes to improved mechanical strength and density by forming solid bridges between particles (Azargohar et al., 2019).

The physical and chemical properties of the feedstock may be altered before pelleting through suitable pretreatment, including grinding, preheating/steam conditioning, and steam explosion (Tumuluru et al., 2011). Grinding contributes to the partial breakdown of lignin, increases the specific surface area, and improves binding, thereby enhancing the mechanical properties of pellets. Additionally, it can promote subsequent bioconversion through size reduction (Chen et al., 2022b; Rijal et al., 2012). Preheating is widely used to improve pellet quality and reduce energy consumption, but it may have an adverse effect on pellet sugar yield (Gong et al., 2019). Conversely, steam conditioning before pelleting with added steam enhances the binding of particles in the pellet die by softening lignin and making it more accessible during pelleting (Takada et al., 2020), while also boosting the bioconversion by disrupting the lignocellulosic structure (Shi et al., 2013). Similar effects can be achieved through the use of steam explosion (Takada and Saddler, 2021; Wu et al., 2022) or ammonia fiber expansion (AFEX) (Bals et al., 2014; Sundaram and Muthukumarappan, 2016a, 2016b).

3.3. Chemical and structural characteristics of biomass pellets

3.3.1. Chemical composition

Pelleting plays a significant role in modifying the physical properties of

			Pellet attributes				
			Moisture	**Density**	**Durability**	**Percent fines**	**Calorific value**
Process variables		Temperature	↑ at moderate (lignin softening); ↓ at excessive (drying/cracking)	↑ (better plasticization and compaction at optimal T)	↑ within Tg; ↓ outside Tg (fracturing, charring)	↑ if overheating → cracks/fines	Little direct effect; may ↑ if drying improves energy density
		Pressure	↑ retention (capillary binding); ↓ at very high (fracturing)	↑(compaction of particles)	↑ (better interlocking); ↓ if too high (brittle fracture)	↓ (less fines at optimal pressure)	Minor ↑ (higher unit density pellets)
		Retention or hold time	Slight ↑ (longer time = moisture redistribution)	↑ (more uniform compaction)	↑ (stress relaxation improves bonding)	↓ (fewer cracks)	No significant effect
		Die geometry (e.g., L/D ratio)	↓ (narrow/small dies increase shear drying).	↑ with smaller diameter & higher compression ratio	↑ (proper L/D ratio gives strong pellets).	↓ at optimal; ↑ at excessive	Neutral (no compositional change)
		Speed (roller/feed rate)	↓ (higher shear rate promotes drying).	↑ at optimal; ↓ if too fast (incomplete compaction).	↑ moderate; ↓ high (cracks, short residence)	↑ at high speeds (fragmentation).	Neutral.
Feedstock variables		Moisture content	↑ at ~8–12% (plasticization); ↓ >20% (puffing, microbial risk)	↑ at optimal (8–12%); ↓ if too wet (>15–20%)	↑ at optimal; ↓ if too dry (<5%) or too wet (>20%)	↑ if too high or too low moisture	↓ if excessive water lowers net heating value
		Particle size, shape, distribution	↑ small; ↓ very fine (dust/poor flow)	↑ with smaller, uniform particles	↑ uniform; ↓ coarse/hetero	↓ uniform; ↑ fines/coarse	Minimal effect; fine particles may slightly ↑
	Biomass composition	Starch	↑ binding via gelatinization	↑	↑	↓	Slight ↑
		Protein	↑ plasticization/binding at elevated T	↑	↑	↓	Neutral
		Lipid/Fat	Neutral to ↓ (lubricating effect reduces bonding)	↓	↓	↑	↑ (higher energy content)
		Cellulose	Neutral (structural backbone)	Neutral	Neutral	Neutral	↑ (structural carbohydrate energy)
		Hemicellulose	Contributes to binding via softening	↑	↑	↓	Neutral
		Lignin	↑ binding when softened (>140 C)	↑	↑	↓	↑ (aromatic content raises calorific value)
		Binders	↑ water retention	↑	↑	↓	Neutral to slight ↑

Fig. 3. Relationships between pellet attributes and densification system variables (Tumuluru et al., 2011; Tumuluru, 2020b).

lignocellulose, making it easier to transport, store, and process. These properties, along with the chemical composition, are crucial for successful bioconversion, including pretreatment and enzymatic saccharification. The pelleting process is always accompanied by a vigorous shearing force and elevated temperature originating from frictional heating, which can partially destroy the intricate architecture of plant cell walls, analogous to mechanical and thermal processing (Guragain et al., 2013; Shi et al., 2013). An overview of published data indicates that pelleting does not significantly alter biomass cell wall composition, although a certain degree of carbohydrate reduction may occur under specific pelleting conditions and feedstock properties (Fig. 4). For example, slight decreases in hemicellulose were noticed in corn stover (Li et al., 2014; Nahar and Pryor, 2017) and softwoods (Kumar et al., 2012) after pelleting. The decrease of hemicellulose could be partially attributed to its higher sensitivity to thermal degradation relative to the other components in the biomass, resulting in degradation during pelleting. Additionally, the relatively abundant moisture in the raw material, when subjected to high temperatures and pressures, generates steam, which also favors hemicellulose degradation (Li et al., 2014).

3.3.2. Structural modifications

While the bulk composition of biomass remains largely unchanged after pelleting, improvements in physical attributes (e.g., bulk density, durability) are accompanied by significant structural alterations at the particle and cell wall levels that directly affect biomass recalcitrance and subsequent biochemical conversion (Fig. 5). Mechanical shear, intensive mixing, and frictional heat during pelleting can disrupt the cell wall structure and reduce biomass particle size (Theerarattananoon et al., 2012; Chen et al., 2023a). Compression of biomass particles in the pellet mill may also induce pore collapse (Bals et al., 2014). In addition, the high temperature and pressure of pelleting have the potential to cause moisture within biomass to generate steam, effectively acting as a mild thermochemical pretreatment (Li et al., 2014). This process can disrupt plant cell wall integrity, leading to cleavage of interunit bonds (e.g., β-O-4′ linkages) in lignin (Wu et al., 2020), alteration of hemicellulose stability (Gong et al., 2021b), and relocation of components within the lignocellulosic matrix, such as lignin (Kaliyan and Morey, 2010).
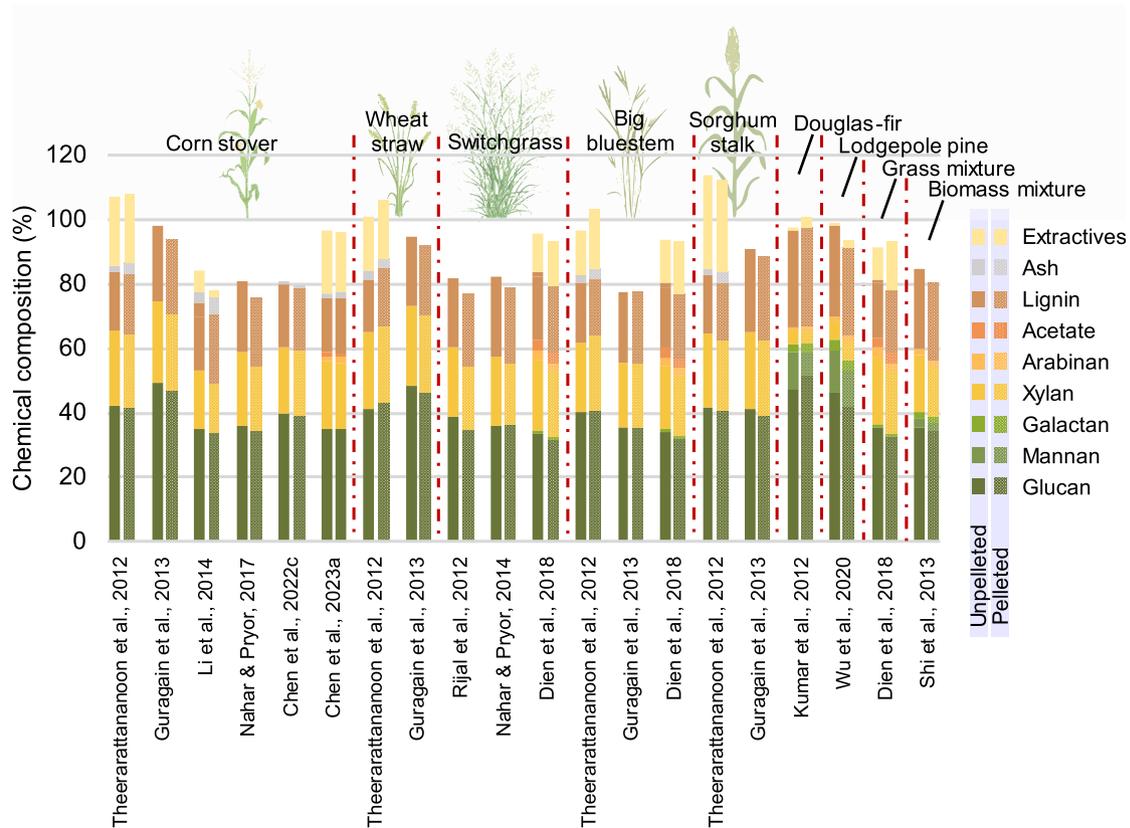
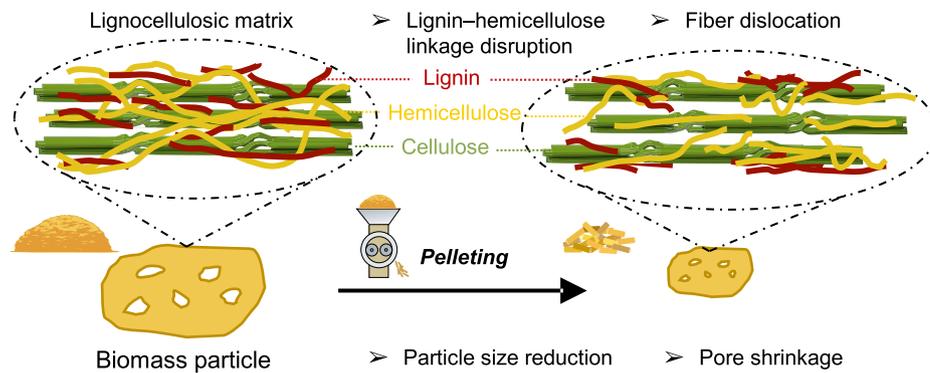**Fig. 4.** Chemical composition of various feedstocks before and after pelleting.

Fig. 5. Schematic representation of structural changes in lignocellulosic biomass during pelleting.

4. Enzymatic saccharification of biomass pellets

4.1. Effect of pelleting on direct enzymatic digestion (non-pretreated biomass)

The suitability of biomass pellets for biochemical conversion is often questioned due to the perceived negative influence of pelleting on enzymatic digestibility, which arises from the diminished substrate accessibility caused by material densification (Kumar et al., 2012). Encouragingly, in the majority of cases, pelleting tends to enhance enzymatic digestibility to varying extents across different biomass species (Fig. 6). This enhancement

could stem from the pelleting process acting as a mild thermochemical pretreatment, which alters the physicochemical characteristics of biomass and reduces its recalcitrance (Fig. 5). In addition, the physical attributes of pellets would also account for the improved enzymatic digestibility, including better mixing facilitated by more free water with reduced pellet dispersion during the initial liquefaction stages (Bals et al., 2014; Serra et al., 2022), as well as particle size reduction resulting from shearing force during pelleting (Bals et al., 2014; Chen et al., 2022b). On the other hand, certain physical alterations, like the reduced water retention caused by the slow release of biomass particulates from the pelleted form and the formation of hardened structures, may diminish enzymatic digestibility. For

instance, a slight impairment in fermentable sugar release was observed in pelleted switchgrass compared to its non-pelleted counterpart (**Fig. 6**), likely due to the incomplete disintegration of switchgrass pellets within 72 h of enzymatic digestion (Rijal et al., 2012). Considering the paramount significance of the physical attributes of pellets in terms of transportation, storage, and handling, an inherent trade-off between these attributes and the enzymatic digestibility of pellets should be thoroughly assessed, in conjunction with other key factors such as energy consumption, to achieve an optimal balance of economic profitability and sustainability.
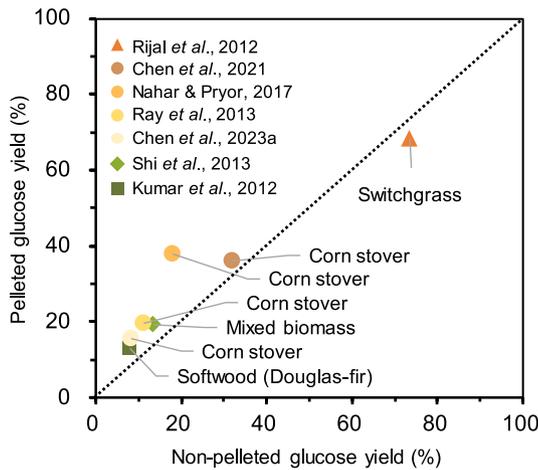

Fig. 6. Effect of pelleting on glucose release through enzymatic hydrolysis of non-pretreated biomass.

4.2. Effect of pelleting on pretreatment and enzymatic digestion

The processes by which lignocellulolytic enzymes hydrolyze polysaccharides in lignocellulose are a function of both substrate reactivity and enzyme activity (**Fig. 7a**) (Ladisch et al., 1983; Chandra et al., 2008). Highly active enzymes are a prerequisite for effective lignocellulose digestion, which is also inextricably linked to various system characteristics conjunctive with the substrate, such as the robustness of enzyme-substrate binding, enzyme component segregation *via* substrate diffusion, as well as its activity loss during the reaction. The nature of lignocellulose itself is also not negligible and affects the digestion at multiple levels, encompassing the microfibril (e.g., degree of polymerization and crystallinity), fibril (e.g., lignin content and distribution), and fiber (e.g., pore size, distribution, and surface area availability) (Esteghlalian et al., 2001).

General biochemical conversion pathways require pretreatment to alter the physicochemical properties of lignocellulose, enabling enzymes to access intricate carbohydrates within and liberate fermentable sugars. A variety of promising pretreatment techniques exists, including dilute acid, steam explosion, liquid hot water, lime, and ammonia pretreatments, which effectively modify the feedstock's composition and structure, thereby enhancing substrate reactivity (Mosier et al., 2005; Chundawat et al., 2011). As depicted in **Figure 7b**, dilute acid pretreatment hydrolyzes hemicellulose and also reduces the lignin content, thereby enhancing cellulose accessibility (Pu et al., 2013; du Pasquier et al., 2023). Liquid hot water pretreatment, also termed as hydrothermal pretreatment (Kim et al., 2009; Chen et al., 2022a), resembles dilute acid pretreatment but uses increased water dissociation at high temperature (Yan et al., 2016) and organic acids liberated from the biomass (e.g., acetic acid from acetate groups) (Mosier, 2013; Pu et al., 2013) to generate acidity without any extra addition of chemicals. An alternative acidic method is the steam explosion pretreatment (Ruiz et al., 2021; Yu et al., 2022b), which involves pressurized steam/water treatment (autohydrolysis) followed by a sudden release of pressure, effectively opening up the feedstock matrix and disrupting the structured fibrous alignment.
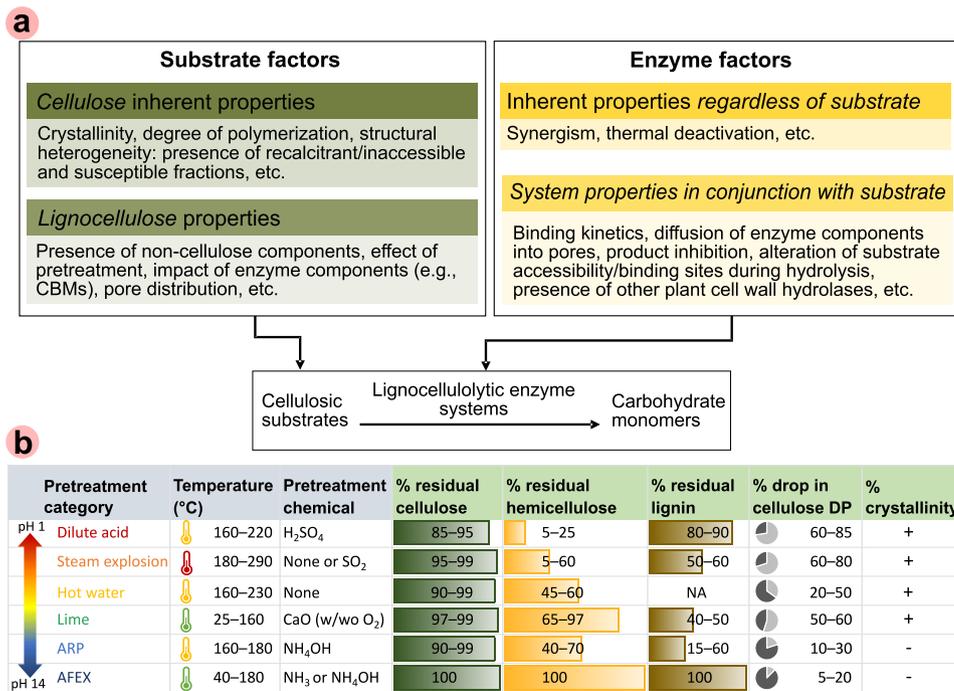
Fig. 7. Factors determining enzymatic digestibility of lignocellulosic biomass and impacts of pretreatment. **(a)** Effects of enzyme and substrate properties on enzymatic digestion of lignocellulosic substrates. Adapted from Chandra et al. (2008). **(b)** Features of leading pretreatments using corn stover as feedstock. Adapted from Chundawat et al. (2011). *Abbreviations:* ARP: ammonia recycle percolation; AFEX: ammonia fiber expansion; DP: degree of polymerization; NA: not available; + or – signs represent the relative augmentation or reduction, correspondingly, in cellulose crystallinity compared to the untreated control.

On the other hand, alkaline pretreatments dissolve lignin and facilitate deacetylation while minimally affecting cellulose content. For example, aqueous alkaline pretreatment using dilute NaOH is a relatively low-severity treatment with high efficiency (Qu et al., 2017). Aside from NaOH-based techniques, alkaline pretreatments utilizing ammonia, such as AFEX (Zhong et al., 2009), solubilize or redistribute lignin while preserving carbohydrates, with the added benefit of easy ammonia recovery due to its high volatility. AFEX combines features of both steam pretreatment and NaOH pretreatment, involving the reaction of moist/wet biomass with ammonia under high pressure to induce ammonolysis and hydrolysis of lignin–carbohydrate complexes and ester linkages, followed by an abrupt pressure release to evaporate the ammonia and open up the biomass structure. While AFEX itself does not fractionate the components of biomass, it disrupts the crystalline cellulose structure and relocates lignin and hemicellulose within the pretreated solids, thereby enhancing cellulose reactivity and enzyme accessibility (Holtzapple et al., 1991). Another ammonia-based pretreatment is ammonia recycle(d) percolation (ARP), which continuously extracts lignin and hemicellulose with an aqueous ammonia solution, resulting in significant delignification and decreased cellulose crystallinity (Kim et al., 2003).

In addition to acidic and alkaline pretreatments, mechanical processes like ball milling (Liu et al., 2019; He et al., 2023; Ding et al., 2025) and disk refining (Chen et al., 2016) are capable of reducing particle size and cellulose crystallinity while increasing the specific surface area and decreasing the degree of Ullah polymerization. Other emerging, milder pretreatment approaches, including the use of organic acids (Lyu et al., 2021, 2022), ionic liquids (Brandt et al., 2013), and deep eutectic solvents (Ullah et al., 2023), have also shown potential for effective lignocellulosic biomass deconstruction under less severe conditions.

The efficiency of any given pretreatment is intricately tied to the distinctive characteristics of lignocellulosic biomass, which are expected to be altered by the pelleting process involving heat and pressure encountered, potentially leading to irreversible pore collapse, reduced fiber permeability, and alterations in lignin melting and surface distribution (Kumar et al., 2012). The impacts of pelleting on the efficiency of pretreatment and subsequent enzymatic saccharification have been examined across a range of feedstocks, including herbaceous, hardwood, and softwood (**Fig. 8**).

Despite employing diverse pretreatment techniques, including dilute acid, aqueous ammonium, and ionic liquids, pellets originating from various agricultural and forestry residues principally exhibited enhanced glucose and/or xylose yield compared to their non-pelleted counterparts, although the hydrolysis yields showed minor variations, with a limited number of studies reporting no impact or a slight decrease in sugar release.

Pelleting demonstrates no detrimental influence on the efficiency of dilute acid pretreatment and, in fact, facilitates greater dissolution of hemicellulose, thereby enhancing the digestibility of pretreated material (Ray et al., 2013; Gong et al., 2021b). The effects are influenced by pelleting parameters and feedstock types (Theerarattananoon et al., 2012). Pelleting parameters, e.g., hammer mill screen size and die thickness, have distinct impacts on glucan and xylan content in dilute acid pretreated biomass (corn stover, sorghum stalk, wheat straw, big bluestem), with the die thickness positively affecting glucan content and negatively affecting xylan content, while screen size has the opposite effect, and lignin content is not significantly impacted (Theerarattananoon et al., 2012). A larger hammer mill screen size recovered more cellulose from corn stover and wheat straw, but less from sorghum stalk and big bluestem, following pretreatment. In contrast, thicker dies had the opposite effect on the different biomass types. Increasing the die thickness (from 31.8 mm to 44.5 mm) improves sugar yield, particularly in corn stover and big bluestem, by enhancing glucan content. A larger hammer mill screen size also contributes to a slight increase in sugar yield. Higher sugar yields in pellets, regardless of conditions and biomass types, suggest that pelleting serves as a preliminary pretreatment step, facilitating the opening of biomass structure before dilute acid treatment.

For acid (SO₂)-catalyzed steam pretreatment of softwood, pelleting may also result in a slight loss of hemicellulose due to the reduced moisture content and smaller particle size. However, it improves the hydrolysis potential of pretreated cellulose, resulting in comparable overall sugar recovery (Kumar et al., 2012). Similarly, liquid hot water (hydrothermal)

pretreatment of biomass pellets results in lower solid recovery, accompanied by a greater removal of lignin and the deconstruction of insoluble carbohydrates into soluble products (Li et al., 2014).

Notably, pelleting actually shows an improved response to alkali pretreatment. Aqueous NaOH pretreatment yields lower solid recovery for pellets compared to unpelleted biomass, primarily due to the increased removal of lignin and the degradation of insoluble carbohydrates into soluble products (Li et al., 2014). Although higher removal of the non-carbohydrate fraction of pellets facilitates the enzymatic hydrolysis of pretreated material, the overall sugar release might not experience a noteworthy improvement in pelleted biomass owing to a greater loss of carbohydrates during the pretreatment. Therefore, a strategy to fully utilize diverse byproducts is desirable for better economic viability (Guragain et al., 2013). Moreover, the impact of pelleting on mass recovery and delignification following alkali pretreatment varies among biomass types. For example, pelleting significantly decreased mass recovery after NaOH pretreatment in corn stover, sorghum stalk, and wheat straw, but not in significant bluestem biomass. Pelleting led to substantial lignin reduction in corn stover and wheat straw, but not in sorghum stalk and big bluestem, while there was no significant variation in glucan and xylan contents between samples before and after pelleting, except for a diminished glucan content in pelleted sorghum stalk following pretreatment (Guragain et al., 2013).

Similar to NaOH-based pretreatment, pelleting biomass enhances the performance of soaking in aqueous ammonia (SAA), thereby improving sugar release by thermally softening or plasticizing lignin in the pelleted biomass (Rijal et al., 2012). Such modification of lignin structure enables milder pretreatment conditions, reduced enzyme loadings, and potential cost savings in the use of pelleted biomass, such as pelleted corn stover, by allowing higher solid loadings without compromising glucose yields and facilitating increased delignification during pretreatment (Nahar and Pryor, 2017). The processing benefits are more pronounced at lower pretreatment severities (Nahar and Pryor, 2017; Pandey et al., 2019). In light of the costly nature of thermochemical pretreatment for lignocellulosic biomass, pelleting presents itself as a viable alternative or a means to mitigate the severity of any thermochemical pretreatment (Rijal et al., 2012).

Other pretreatments, such as ionic liquid (IL) pretreatment, confirm that pelleting has no negative impacts on pretreatment and subsequent enzymatic hydrolysis, where pellets from blended biomass (equal masses of switchgrass, lodgepole pine, corn stover, and eucalyptus) showed comparable sugar yields to the mixed flour format, and despite the denser nature of the pellets, they exhibited similar swelling and solubilization behavior during IL pretreatment as the flour, resulting in similar feedstock compositions (Shi et al., 2013).

Multiple characterization techniques have been employed to identify the factors contributing to the pretreatment effectiveness and sugar yields of pelleted biomass, including shearing and heating during the pelleting process, which modify the biomass's macrostructure and disrupt lignin-hemicellulose linkages (Ray et al., 2013; Nahar and Pryor, 2017). Scanning electron micrographs (SEM) reveal distinct morphological differences between pelleted corn stover and non-pelleted materials, with rougher surfaces and increased surface area observed in the pelleted material, which may potentially enhance bioconversion (Ray et al., 2013; Nahar and Pryor, 2017). Brunauer–Emmett–Teller (BET) nitrogen adsorption indicates that pelleting reduces the surface area and pore volume, but increases the average pore size; however, these differences become insignificant after dilute-acid pretreatment (Ray et al., 2013). X-ray diffraction (XRD) indicates no significant alteration of microcrystalline cellulose due to pelletization (Shi et al., 2013). FTIR spectra suggest the cleavage of acetyl and uronic ester linkages in hemicellulose and/or lignin during pelleting (Nahar and Pryor, 2017).

The influence of pelleting on pretreatment and enzymatic digestion is closely linked to the physicochemical modifications induced during the densification process. Despite little to no change in biomass composition, structural alterations, such as reduced particle size and modifications to hemicellulose and lignin, can facilitate pretreatment and improve cellulose accessibility (**Fig. 7b**). In contrast, pore shrinkage resulting from hornification might diminish these benefits, limiting pretreatment efficiency and hydrolysis performance. Overall, the impact of structural modifications

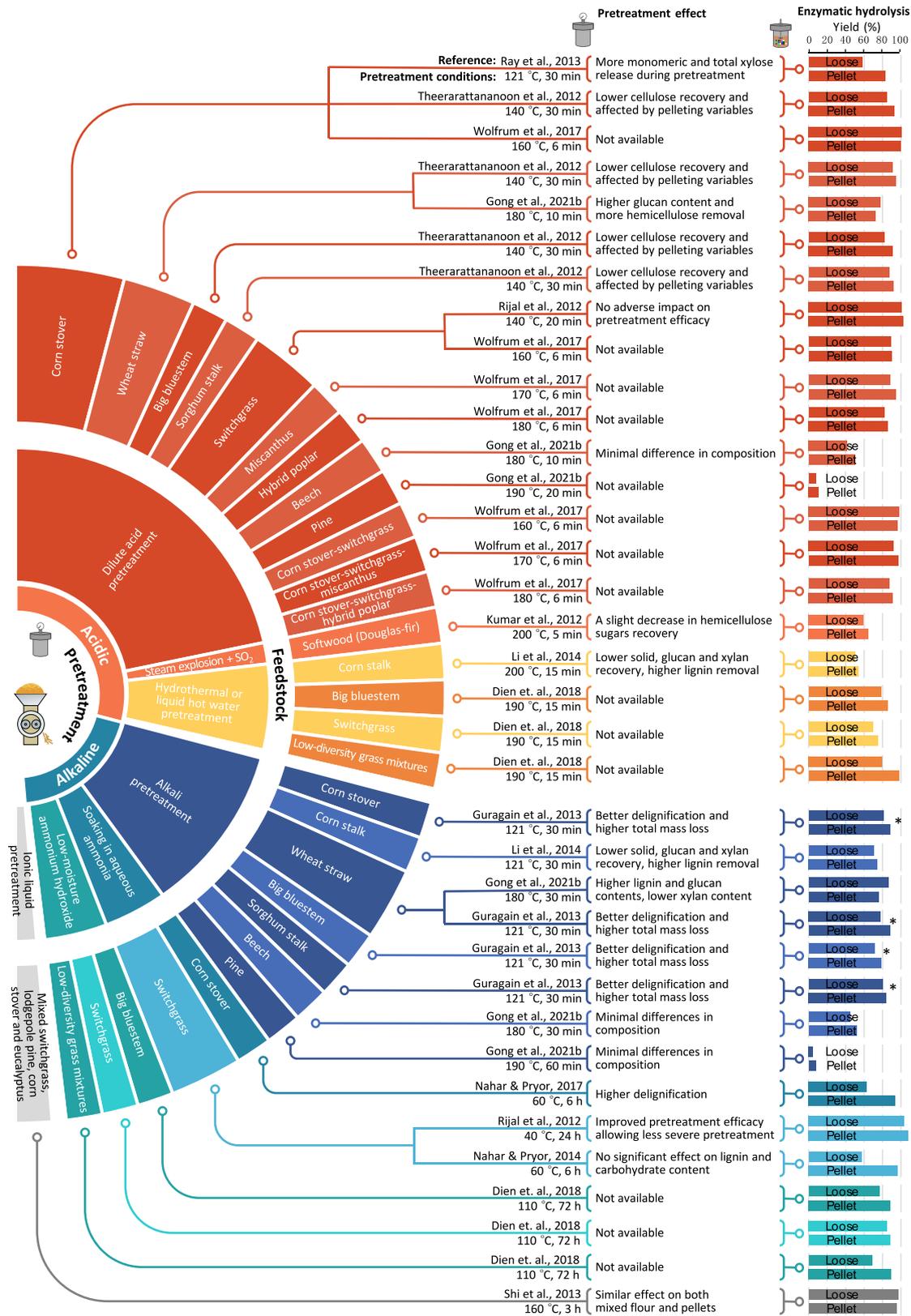**Fig. 8.** Overview of pelleting impact on pretreatment and enzymatic hydrolysis of lignocellulosic biomass. The yield values represent glucose yield, except for those marked with *, which include both glucose and xylose yields.

induced by densification on biochemical conversion outcomes is determined by the interplay of biomass characteristics, densification technology, and pretreatment type and severity.

4.3. Pelleted biomass enzymatic saccharification scenarios

To support transportation, storage, and processing while lowering associated costs, biomass can be pelleted and then delivered to lignocellulosic biorefineries for pretreatment and subsequent conversion (Dien et al., 2018; Chen et al., 2022c). Pelleting can also be performed following pretreatment, considering that the pretreated biomass is more sensitive to the pelleting process than the untreated materials, primarily due to lignin alterations (Bals et al., 2014; Wu et al., 2022). Moreover, biomass pelleting and pretreatment can be carried out simultaneously by densifying the biomass with chemicals, such as alkalis (Chen et al., 2021), acids (Yuan et al., 2022), and other additives (Gong et al., 2021a) (Fig. 9). This integrated process not only simplifies processing steps and reduces the corresponding expense but also produces highly dense and durable pellets with excellent resistance to microbial contamination and superior enzymatic digestibility (Chen et al., 2021). Different processing schemes have distinct impacts on enzyme-mediated conversion as well as overall costs. Therefore, a better understanding of the features of various pelleting strategies is critical for efficient and cost-competitive process design optimization.

4.3.1. Pelleting of non-pretreated feedstocks

Pelleting biomass to facilitate transportation, storage, and handling provides varying degrees of additional benefits to pretreatment and enzymatic hydrolysis (Fig. 8). Besides reducing pretreatment severity and enhancing pretreatment efficiency, pelleting enables high solids loading for both pretreatment and enzymatic hydrolysis, resulting in sugar-rich syrups and cost benefits in terms of both capital and operating expenses, thereby increasing the overall economic viability (da Silva et al., 2020). However, the mass/heat transfer during pretreatment should be considered to determine appropriate conditions, prevent excessive financial burden, and minimize undesired product formation (Chen et al., 2021). Given the variability in the effectiveness of pretreatment methods across different biomass types, it is essential to individually optimize the pelleting procedure for each specific biomass type, utilizing a favorable pretreatment approach. Therefore, conducting a comparative study that examines various pretreatment techniques and pelleting variables to assess their combined impact on biomass delignification for enhanced enzymatic hydrolysis is recommended for future research (Guragain et al., 2013).

4.3.2. Pelleting of pretreated feedstocks

Pre-steaming or steam explosion before pelleting is advantageous for activating lignin, potentially by lowering its glass transition temperature through interaction with water at high temperatures during densification, thereby improving pellet durability as lignin serves as a binder (Tumuluru, 2020b). In a relevant study, pelleting SO_2-catalyzed steam-pretreated softwood chips decreased hydrolysis yields from 64% to 53%. However, this reduction could be restored by mechanically disintegrating the pellets after overnight water soaking, thereby regaining the hydrolysis yields to their original levels achieved with steam-treated chips (Kumar et al., 2012). Furthermore, incorporating lignosulfonate before pelleting improves pellet durability while facilitating subsequent hydrolysis (Takada and Saddler, 2021). Collectively, steam pretreatment relocates the lignin in the plant cell wall to the fiber surface, thereby enhancing particle binding and resulting in a significant improvement in pellet durability, which alleviates the necessity


I Traditional, using loose biomass for each step

Harvest & Baling → Transportation → Storage → Milling → Pretreatment → Enzymatic hydrolysis
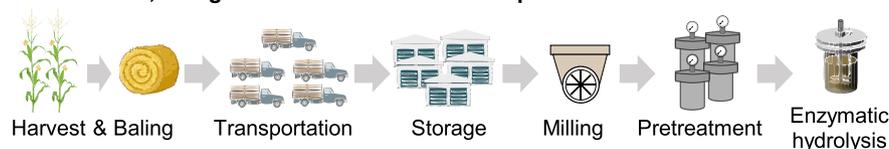
Issues:
 i : High transportation & storage cost
 ii : Biomass contamination by microbe
 iii : Low working mass during pretreatment
 iv : Severe pretreatment conditions: high cost & toxic degradation products

II Densification before transportation

Harvest & Densification → Transportation → Storage → Pretreatment → Enzymatic hydrolysis
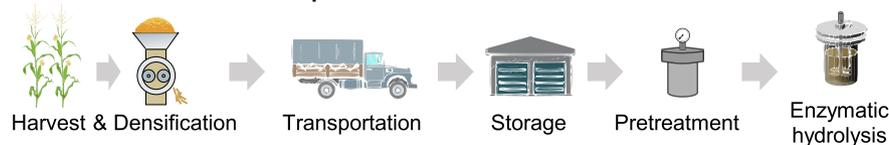
Advantages:
 Reduced transportation & storage cost
Issues:
 i : Biomass contamination
 ii : Mass transfer & heat transfer during pretreatment
 iii : Severe pretreatment conditions: high cost & toxic degradation products

III Pretreatment and densification before transportation

Harvest & Milling → Pretreatment → Densification → Transportation → Storage → Enzymatic hydrolysis
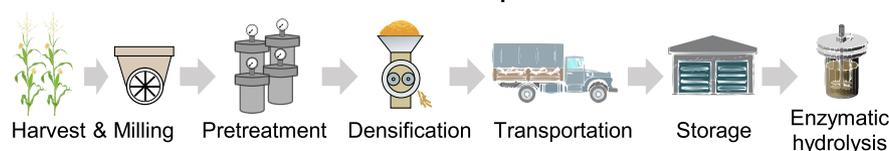
Advantages:
 i : Reduced transportation & storage cost
 ii : Reduced contamination
Issues:
 i : Low working mass during pretreatment
 ii : Severe pretreatment conditions: high cost & toxic degradation products

IV Pretreatment integrated with densification, transportation and storage

Chemicals
Harvest & Densification → Transportation → Storage → Autoclave (optional) → Enzymatic hydrolysis
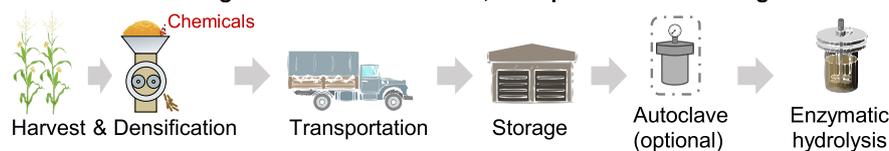
Advantages:
 i : Reduced transportation & storage cost
 ii : Contamination prevented
 iii : Low cost pretreatment occurring during densification, transportation and storage
 iv : Mild further treatment conditions with high working mass if needed
 v : Fewer toxic degradation products

Fig. 9. Various scenarios for bioconversion of lignocellulosic biomass. Adapted from Chen et al. (2021).

for size reduction (Kumar et al., 2012). Negative impacts from drying and pelleting can be alleviated by incorporating a neutral sulfonation or alkaline deacetylation step before pre-steaming (Tang et al., 2018). Further enhancement in hydrolysis can be achieved by following a mild mechanical refining process (Wu et al., 2022).

As an alternative means to fluidize and relocate feedstock lignin, AFEX has shown promise in pretreatment for improving pellet quality without generating a separate liquid stream. However, the hornification of pelleted biomass resulting from pressing and drying during pelleting leads to decreased enzymatic hydrolysis. Regarding AFEX-pretreated corn stover, pelleting can enhance enzymatic hydrolysis at high solid loadings by reducing particle size and improving mixing in enzymatic hydrolysis slurries, which is attributed to the lower water absorption and retention of pellets compared to unpelleted stover (Bals et al., 2014). Sundaram and Muthukumarappan investigated the impact of pelleting conditions and biomass blending on the physical attributes of pellets and enzymatic digestibility, revealing that pelleting and biomass blending affect the compaction characteristics but do not significantly influence the sugar release from AFEX-pretreated corn stover and switchgrass (Sundaram and Muthukumarappan, 2016b). Additionally, they observed that AFEX pretreatment improved the bulk density and hardness of biomass pellets, while the glucose and xylose yields remained unaffected by the hammer mill screen size and extrusion temperature (Sundaram and Muthukumarappan, 2016a). Moreover, coupling AFEX-treated pellets with a CIII$_I$-activation step using anhydrous liquid ammonia at room temperature can significantly reduce the enzyme dosage required for efficient high-solids enzymatic hydrolysis by converting the native crystalline cellulose I$_\beta$ (CI$_\beta$) to more digestible allomorph cellulose III$_I$ (CIII$_I$) (Mokomele et al., 2022).

Pretreatments such as pre-steaming/steam explosion and AFEX prior to pelleting promote pellet durability, facilitating improved transportation and handling, as well as resistance to microbial contamination during storage. However, the loose material treated to pretreatment before pelleting is less effective than pellets at high solids loading. Moreover, the negative impact of pelleting pretreated biomass on bioconversion should also be considered for assessing overall economic feasibility.

4.3.3. Pelleting with chemicals

The addition of surfactant (polyethylene glycol (PEG) 6000) before pelleting has been shown to have beneficial impacts on the pelleting process and enzymatic saccharification. Surfactant-assisted pelleting of wheat straw and pine reduced friction during pelleting and specific energy consumption, while also facilitating enzymatic hydrolysis following acid or alkaline pretreatment. This enhancement in sugar yield can be attributed to the ability of PEG 6000 to mitigate the breakage of β-O-4 bonds during pretreatment, thus inhibiting the generation of phenolic hydroxyl groups that can impede enzyme activities (Gong et al., 2021a).

Densifying lignocellulosic biomass (DLC) with alkaline or acidic chemicals is another promising process that combines biomass densification and pretreatment into a single step. It improves biomass logistics and favors enzymatic hydrolysis without the need for additional pretreatment, or with only a general autoclave treatment, thereby avoiding the high temperatures and high pressures common in traditional pretreatments. Densifying corn stover with alkaline chemicals yielded high-density and durable pellets, which preserved sugars and prevented microbial contamination. Pellets containing calcium hydroxide achieved an ethanol yield of 21.4 g per 100 g of corn stover, and this yield was further improved to 25.3 g per 100 g of biomass after undergoing a regular autoclave pretreatment. Remarkably, a satisfactory ethanol titer of up to 70.6 g/L was attained in the absence of washing or detoxification steps (Chen et al., 2021). Similarly, this easy-to-implement process facilitates achieving a high ethanol titer of 68.1 g/L for corn stover, pelleted with sulfuric acid and subjected to regular steam autoclave treatment (Yuan et al., 2022). Mechanical densification facilitates acid penetration into the biomass matrix, enhancing xylan accessibility to acid catalysis and thus effectively disrupting the biomass structure by solubilizing xylan at low temperatures (Jiang et al., 2023; Yuan et al., 2023). High enzymatic digestibility and fermentability were also observed for sugarcane bagasse pellets densified with sulfuric acid, resulting in an ethanol titer of up to 77.51 g/L after an autoclave treatment followed by

simultaneous saccharification and co-fermentation at high gravity (30% solid loading). This achievement of a high ethanol titer, reaching nearly 10% (v/v), is desirable without the need for washing or detoxification (Shen et al., 2022). Corncob and rice straw treated similarly reached even higher ethanol yields of 82.0 g/L (Liu et al., 2022) and 82.9 g/L (Yuan et al., 2023), respectively, at a 35% solid loading. Coupling metal salts with sulfuric acid in a binary DLC system offers benefits beyond superior sugar production (over 90% enzymatic sugar yield and a fermentable sugar concentration of 212.3 g/L), including the generation of highly adsorptive bio-adsorbents from biomass residues effective for dye wastewater treatment (Yuan et al., 2024). The DLC process has also been extended to integrate deep eutectic solvents (DES), which are effective in biomass fractionation but face challenges in industrial applications due to the high dosage of DES and low biomass loading. Using a limited amount of DES in densification pretreatment triggers efficient lignocellulose fractionation and bioconversion. A DES dosage of 2 g/g biomass delignified 75% of corn stover while preserving over 95% of the cellulose at 50% solid loading. This approach yielded superior bioconversion efficiency, with a maximum total fermentable sugar concentration exceeding 300 g/L and an ethanol concentration reaching 110 g/L, achieving over 90% glucan conversion even at a 35% solid loading during enzymatic hydrolysis (Shen et al., 2023).

Several factors contribute to the efficiency of densifying lignocellulosic biomass with chemicals, followed by regular steam autoclaving (DLCA). For instance, controlling ash content is important for optimizing this process. High ash content limits the efficiency of pretreatment and enzymatic digestibility with sulfuric acid but enhances them with calcium hydroxide, as ash anions neutralize the acid while synergizing with the alkali (Sha et al., 2024). Storage conditions also have varying influences on the enzymatic digestibility and fermentability of pellets derived from densifying lignocellulosic biomass with chemicals, with corn stover pellets containing calcium hydroxide showing enhanced enzymatic digestibility with the increase of storage temperature and time, whereas pelleted corn stover incorporating sulfuric acid is sensitive storage conditions and highest the ethanol titer was achieved by storing at ambient temperature for two weeks (Wang et al., 2022). Moreover, feedstock selection is an unignorable factor influencing the process performance and economics of DLCA. A techno-economic analysis (TEA) of different crop straws in cellulosic ethanol production using DLCA pretreatment with calcium hydroxide shows that corn stover, rice straw, and wheat straw have minimum ethanol selling prices (MESP) of 2.21 USD/gal ethanol, 2.12 USD/gal ethanol and 2.03 USD/gal ethanol, respectively, at a 2000-tonne-per-day biorefinery with a 10 % internal rate of return. Notably, their mixture results in an MESP of 2.02 USD/gal ethanol, highlighting the potential for large-scale production (Yu et al., 2024). Economic analysis of DLCA indicated that the savings in feedstock logistics alone nearly compensate for the cost of this treatment itself (Yu et al., 2022a).

5. Practical considerations

Despite the numerous advantages of pelleted biomass for storage, processing, transportation, and conversion, current biorefineries face challenges in adopting biomass pellets as feedstock due to the high costs associated with producing pelleted biomass using existing densification methods. Traditional pelleting methods require stringent moisture management and a feedstock with a moisture content between 10% and 15% wt%. Such moisture constraints require investment in additional rotary drying equipment and relatively expensive pelleting dies. To consider this cost in the context of corn stover as a bioenergy feedstock, we can observe that an analysis performed by Tumuluru and Yancey (2018) suggested a per tonne cost of over USD20 (2018 dollars) and that a recent market estimate placed the value of corn stover at USD40–60 per dry tonne (Edwards, 2020). In comparison, a State of Technology Report published by the Department of Energy (2022) estimates that a per-dry-tonne feedstock cost of USD71 or less, as delivered to the refinery (2017 dollars), is necessary for corn stover to be an economically viable bioenergy feedstock in the current market (U.S. Department of Energy, 2020). As such, investing in capital and operating costs necessary for densification may be difficult.

However, ongoing advancements in milling and pelleting technologies

by industry and national laboratories, such as Idaho National Laboratory (INL), have shown promising results in reducing pellet production costs. Innovative preprocessing techniques, such as INL-designed fractional milling, high-moisture pelleting, and low-temperature drying, have successfully achieved more than a 50% reduction in pellet production costs (**Fig. 10**) (Tumuluru, 2020c). The cost benefits of increased density and stability must be shown to outweigh these costs. Or, as described in this work, additional benefits of densification in conversion yields may further justify densification. Outside of the impact of moisture on densification discussed above, more traditional challenges of feedstock preprocessing, such as those related to feedstock variability, are unlikely to represent significant barriers to the implementation of densification, as densification is typically less impacted by these factors than other necessary feedstock operations, such as size reduction and reactor feeding.
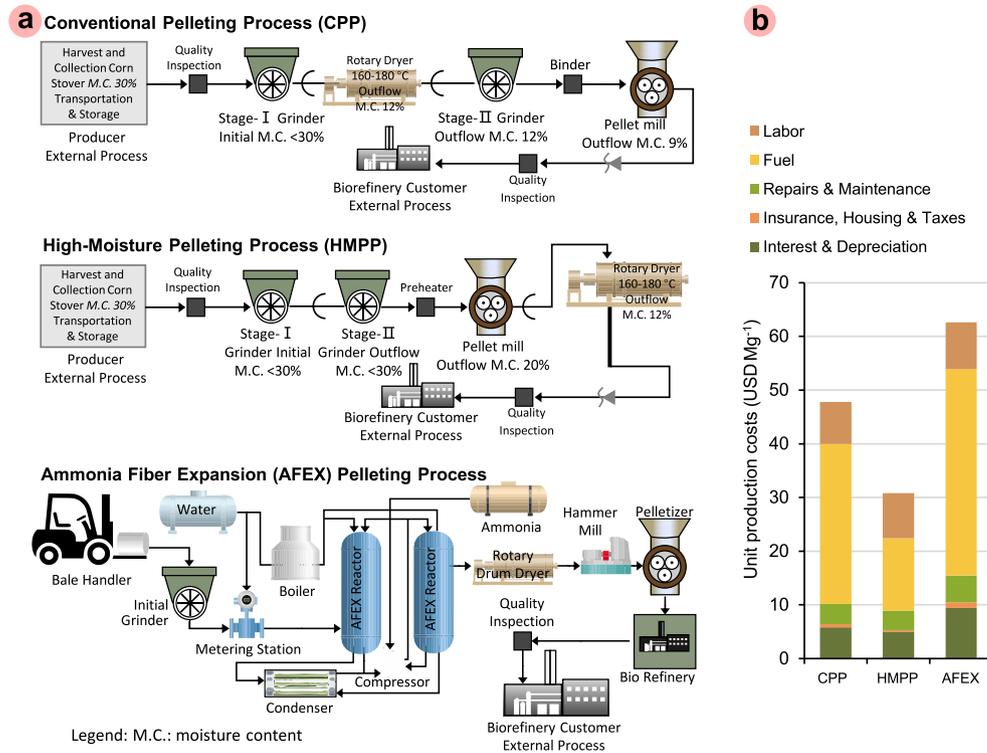

Fig. 10. Techno-economic analysis of decentralized biomass processing depots. **(a)** Three different technical configurations of the depot (adapted from Lamers et al. (2015), Figure 2, p. 209, and Figure 3, p. 210 (Elsevier)). **(b)** A comparison of total cost between depot configurations (adapted from Lamers et al. (2015), Figure 4, p. 211 (Elsevier)).

5.1. Energy consumption

Pelleting is an energy-intensive process that significantly contributes to production costs (**Fig. 10b**) (Lamers et al., 2015). The specific energy required for biomass pelleting depends on machine parameters, process variables, and feedstock characteristics (Tumuluru et al., 2011). Variability in moisture complicates feed handling and increases the costs of preprocessing and drying. Preprocessing, including size reduction and drying, accounts for a substantial portion of energy consumption, with drying consuming more energy than pelleting itself (**Table 3**) (Yancey et al., 2013). Accordingly, efficient moisture management is crucial to reducing pellet production costs. An example of a cost-effective approach involves employing high-moisture pelleting combined with low-temperature drying (**Fig. 10**). This technique involves a short-duration, low-temperature drying process prior to pelleting, rather than the conventional drying step after grinding, making post-pelleting drying optional (Tumuluru, 2014). It offers a cost-effective alternative to biomass drying by partially drying biomass through preheating, generating frictional heat from both grinding and pelleting, and cooling during the pellet production process. If needed, affordable low-temperature dryers, such as grain dryers, can further dry pellets to achieve stable moisture content levels of less than 10% (Kenney et al., 2013). Additionally, fractional milling, which involves increasing the first-stage grinder's screen size and inserting a separator between the first-stage and second-stage grinders to bypass compliant particles, saves energy, avoids redundant grinding, and narrows the particle size distribution (Kenney et al., 2013; Tumuluru and Yancey, 2018). Optimizing the pelleting process through the implementation of these technologies is essential for reducing energy consumption and, consequently, production costs (**Fig. 10**). In commercial-scale unit operations, the energy consumption for high-moisture pelleting was reported to range between 98 and 124 kWh/tonne, representing a 64–72% reduction in energy consumption compared to conventional pelleting, which consumes approximately 350 kWh/tonne to dry biomass from 30% to 10% moisture content before pelleting (Tumuluru, 2023).

Table 3.
Energy consumption of various conventional pelleting unit operations for different biomass materials (Yancey et al., 2013).

Biomass Type	Stage I Grinding (kWh/tonne)	Rotary Drying (kWh/tonne)	Stage II Grinding (kWh/tonne)	Pelletization (kWh/tonne)
Switchgrass	11.72		18.55	45.20
Eucalyptus	94.11	401.78	49.97	124.13
Lodgepole pine	94.11		43.21	48.94
Corn stover	19.38	340.77	34.54	56.43
Four crop mix	54.83	185.64	56.52	76.36

5.2. Economics and sustainability

Considering the significant energy consumption in pellet production, it is crucial to acknowledge and prioritize the downstream financial (Table 4) and environmental (Table 5) advantages associated with pelleted biomass applications, which are often overlooked despite extensive research on the economic and logistics benefits of biomass pelleting (Shahrukh et al., 2016; Manandhar and Shah, 2017). The question of whether pelleting yields net energy and emission savings during feedstock transportation and processing was examined through a life cycle assessment (LCA) encompassing the pelleting, transportation, and SAA pretreatment stages of corn stover. Despite higher greenhouse gas (GHG) emissions from pelleting that surpass transportation levels by a factor of up to 25 in centralized dry densification systems (Nahar et al., 2021), leveraging the benefits of pretreatment processes has the potential to offset and reduce overall emissions. Pelleted biomass subjected to SAA pretreatment significantly reduced energy consumption (56%), allowed higher solid loadings, and thus the decreased demand for chemicals by 56% and water by 49% (Nahar et al., 2021). A comparative TEA and LCA revealed that pelleted corn stover offers superior economic and environmental benefits for ethanol production through SAA pretreatment and enzymatic hydrolysis. Pelleted biomass provides a lower minimum ethanol selling price through shorter processing times, higher solids loadings, and reduced ammonia and enzyme requirements, while also exhibiting a 50% lower life cycle GHG emission than non-pelleted biomass due to reduced chemical needs, thereby offsetting the higher energy consumption during pelleting (Nahar et al., 2021). In contrast, a techno-economic analysis reveals that densified corn stover with anhydrous liquid ammonia pretreatment under mild conditions yields a slightly higher MESP compared to loose AFEX-treated biomass at the same scale; however, its improved logistics and handling could enable larger plants, potentially offsetting these costs (Zhang et al., 2025).

Densifying lignocellulosic biomass with chemicals (calcium hydroxide), followed by a regular steam autoclave treatment, also demonstrates economic advantages in biomass logistics and conversion for the profitable production of cellulosic ethanol. Compared to a biorefinery utilizing dilute acid pretreatment, it exhibited significantly lower feedstock logistics costs, with the savings almost compensating for the densification expenses. Additionally, it improved ethanol production and reduced pretreatment expenditures. For a biorefinery processing 2000 Mg rice straw daily, it resulted in a 17.9% lower total capital investment and a 30.0% lower minimum ethanol selling price compared to the dilute acid pretreatment-based biorefinery (Yu et al., 2022a). The combination of this pelleting strategy with other promising preprocessing technologies, such as fractional milling and high-moisture pelleting, holds the potential to enhance the financial and environmental benefits associated with biomass pellet formation and utilization.

6. Conclusions and future perspectives

Pelleting biomass to improve feedstock logistics brings multiple additional benefits. It tends to enhance the enzymatic digestibility of unpretreated lignocellulose, even without substantial modification in its chemical composition. Pretreatment efficacy is also prone to varying degrees of improvement, depending on the type of biomass, pelleting variables, and the specific type and conditions of the pretreatment process. All these factors contribute to the performance of enzymatic saccharification, thereby determining the overall cost and environmental impact associated with deriving fermentable sugars from lignocellulosic

Table 4.
Survey of recent technoeconomic studies on pelletization of lignocellulosic biomass for biochemical conversion.

Source	Feedstocks	Reference Year	Pretreatment or Process	Feedstock Cost (USD/dry tonne)	Ethanol Yield (gal/dry tonne)	MESP (USD/gal)	Remarks
Pandey et al. (2021)	Corn stover	2017	SAA* (loose; 50 °C, 14 h, 14% ammonia); 25 FPU + 500 XU/g glucan	95	67.0	4.41	Bulky feedstock, prolonged hydrolysis, and high enzyme usage.
			SAA (pelleted; 55 °C, 19 h, 16% ammonia); 5 FPU + 100 XU/g glucan	106	66.7	3.83	Pelletization reduces the severity and enzyme loading, allowing for higher solids.
Morais et al. (2022)	Sugarcane bagasse	2012	COBRA (100 °C, 3.5 h, 1:1 [NH₃:BM (g/g)]); 15 mg of protein/g glucan	77.6-82.1	85.7	1.49	Higher delivered biomass price, good yield, and low cost.
			COBRA-LE (100 °C, 3.5 h, 1:1 [NH₃:BM (g/g)]); 15 mg of protein/g glucan	77.6-82.1	89.0	1.45	Lower enzyme loading, higher yield, and lowest MESP.
			EA (120 °C, 0.5 h, 6:1 [NH₃:BM (g/g)]); 15 mg of protein/g glucan	61.5-71.3	84.3	1.58	Higher ammonia and pressure result in higher costs.
			AFEX (140 °C, 1 h, 1:1 NH₃:BM (g/g)); 25 mg protein/ g glucan	61.5-71.3	86.0	1.46	High yield, higher enzyme requirement.
			StEx (200 °C, 0.1 h); 25 mg protein/g glucan	61.5-71.3	54.4	1.98	Low yield due to sugar degradation.
Zhang et al. (2025)	Corn stover	2012	AFEX (120 °C, 0.5 h, 1:1 NH₃:BM (g/g)); 15 mg protein/ g glucan	75	66.3	1.99	Lower feed cost, lower yield, low fill ratio.
			COBRA (67 °C, 5.5 h, 0.8:1 NH₃:BM (g/g)); 15 mg protein/g glucan	90	72.6	2.25	Higher costs associated with longer residence times; better logistics for larger biorefineries.
Chen et al. (2021)	Corn stover	N.A.	DLCA(ch) (autoclaved 121 °C, 1 h); 10 mg protein/g glucan	59.9	80.3	2.05	Lower pretreatment cost than DA.
			DA (160 °C, 20 min, 0.05:1 H₂SO₄: BM (g/g); 30 mg protein/ g glucan	62.6	~80	2.37	More inhibitors, higher cost.
Yu et al. (2022a)	Rice straw	2017	DLCA(ch) (autoclaved 121 °C, 1 h); 20 mg protein/g glucan	61.8	76.3	2.12	Lower logistics costs, higher yield, and lower pretreatment costs compared to DA.
			DA (160 °C, 20 min, 0.1:1 H₂SO₄: BM (g/g); 20 mg protein/g glucan	61.7	55.6	3.03	Low yield, higher MESP.

*Abbreviations: AFEX: Ammonia Fiber Expansion; BM: Biomass; COBRA: Compacted Biomass With Recycled Ammonia Process; COBRA-LE: COBRA Low Enzyme; DA: Dilute Acid; DLCA(ch): Densifying Lignocellulosic biomass with chemicals under Autoclave; EA: Enhanced Ammonia; FPU: Filter Paper Unit; MESP: Minimum Ethanol Selling Price; SAA, Soaking in Aqueous Ammonia; StEx: Steam Explosion; XU: Xylanase Unit.

Table 5.
Summary of life cycle perspectives of pelleted vs. non-pelleted corn stover for bioethanol production with SAA pretreatment.

	Non-Pelleted	Pelleted
Gate-to-gate (farm gate to end of pretreatment) (Nahar et al., 2021)		
Pretreatment parameters		
Solid loadings (%)	10	20
Temperature (°C)	60	56
Residence time (h)	24	4
Ammonia (%)	15	12
Chemical inputs (kg/Mg biomass)		
Water	~9,300	~4,700
Ammonium hydroxide	~1,350–1,400	~500–550
Sulfuric acid	~220–230	~150–160
Energy Input (kg/Mg biomass)		
Pelleting	0	521
Transportation	147	110
Pretreatment	1681	748
GHG Emission (kg $CO_{2\,eq}$/Mg biomass)		
Pelleting	0	34
Transportation	1.88	1.41
Chemical production	1773	720
Cradle-to-grave (2000 Mg/d biorefinery) (Pandey et al., 2021)		
Pretreatment conditions	50 °C, 14 h, 14% NH_3	55 °C, 19 h, 16% NH_3
Enzyme loading	25 FPU* + 500 XU/g glucan (1:1 FPU:CBU)	
Hydrolysis performance		
Time for 90% glucose yield (h)	33	48
Xylose Yields (%)	72	62
Ethanol Yield (gal/dry tonne biomass)	67.0	66.7
MESP (USD/gal)	4.41	3.83
Emission in kg $CO_{2\,eq}$/MJ of produced ethanol	0.097	0.048

* Abbreviations: CBU: Cellobiase Units; FPU: Filter Paper Unit; MESP: Minimum Ethanol Selling Price.

biomass. Currently, the production of biomass pellets is a highly energy-intensive process, where significant energy is consumed during preprocessing steps, including size reduction, drying, and pelletization. Emerging preprocessing innovations, including fractional milling, high-moisture pelleting, and low-temperature drying, hold promise to lower energy consumption. Optimizing pelleting strategies, such as densifying lignocellulosic biomass with chemicals by combining pelleting and pretreatment into a single process, is crucial to highlight the economic and environmental advantages of pelleted biomass in biorefineries (Table 6).

To fully utilize the potential of lignocellulose for decarbonizing the fuel, chemical, and material industries through pelleting, there is a need for fundamental and technical advancements. A comprehensive understanding of the interplay between feedstock, pelleting, pretreatment, and enzyme-mediated conversion is critical for designing and optimizing not only upstream feedstock preprocessing and pelleting processes but also downstream pretreatment and conversion strategies. Particularly, an in-depth characterization of feedstock features resulting from pelleting, such as lignin attributes that determine pretreatment effectiveness and enzymatic digestibility, is highly desired. Integrating innovative preprocessing techniques and strategies with economic and sustainability analysis is central to the commercialization of biomass pellet production and utilization. In this context, multi-objective optimization (e.g., balancing sugar yield, energy use, costs, and GHG emissions) will be essential for guiding process design and identifying the most sustainable pelleting strategies. Optimization of each step, such as on-farm pellet processing and the use of mobile pelleting machines, enhances practicality and logistics efficiency. Combining pelleting and pretreatment offers attractive prospects; however, careful consideration and optimization of equipment requirements, including corrosion tolerance and overall cost implications, are essential. Other engineering considerations, such as reactor design for biomass pellets, are also crucial to ensure adequate mass/heat transfer with reduced solvent usage. Furthermore, the valorization of lignin is central to the economic feasibility of the lignocellulosic biorefining industry, necessitating an understanding of the responses of pelleting on lignin structure as well as potential applications.

Looking forward, emerging digital tools hold significant promise for optimizing pellet-based biorefineries. Artificial intelligence (AI) and machine learning can support the predictive modeling of pelleting variables and conversion outcomes. Digital twins enable real-time simulation and optimization of integrated operations, while sensor-based control systems provide adaptive monitoring of pellet quality, energy consumption, and enzymatic performance. Incorporating these technologies can accelerate process optimization, reduce experimental costs, and support scale-up toward efficient and sustainable closed-loop biorefinery systems.

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Table 6.
Research gaps and future perspectives for pelletization in biorefineries.

Aspect	Current Challenge	Future Research Directions
Process variables & feedstock diversity	Pellet quality and saccharification vary widely by biomass properties and operating parameters	Develop biomass-specific optimization frameworks, supported by AI/ML models, to predict pellet attributes, energy use, and sugar yields
Digital and smart technologies	Lack of real-time optimization and adaptive control of pelletization and conversion processes	Introduce AI, digital twins, and sensor-based systems for predictive modeling, real-time simulation, and adaptive monitoring of pellet quality, energy use, and conversion performance
Lignin transformation & valorization	Pelleting alters lignin (relocation, plasticization, hornification), but impacts on lignin quality and valorization pathways remain poorly understood	Conduct systematic studies on lignin modifications under diverse pelleting strategies and link them to valorization opportunities (e.g., binders, advanced materials, lignin-derived chemicals)
Advanced pelleting strategies	Conventional pelleting is energy-intensive; separate pretreatment increases costs. Current pelleting often requires size reduction and drying, adding high energy demand.	Integrate chemical-assisted densification to combine pelleting and pretreatment in a single step, while advancing innovations such as high-moisture pelleting, low-temperature drying, and optimized size reduction to lower energy inputs. Evaluate techno-economic and environmental trade-offs of these strategies using TEA–LCA frameworks
Mobile/on-farm pelleting & scale-up integration	Mobile systems exist, but they lack validation at a commercial scale; integration with current biorefineries/agricultural plants remains limited.	Develop pilot- and commercial-scale demonstrations of mobile/on-farm pelleting. Optimize equipment design for energy efficiency and robustness. Explore co-location with existing biorefineries, grain facilities, or agro-industrial plants to reduce capital and accelerate deployment

Author Contributions

Xueli Chen: conceptualization, data analysis, generation of figures and tables, writing (initial draft, review, editing). **John E. Aston:** data interpretation, writing (review and editing). **David N. Thompson:** writing (review and editing). **Michael R. Ladisch:** supervision, conceptualization, data interpretation, writing (review and editing). **Nathan S. Mosier:** conceptualization, supervision, data interpretation, writing (review and editing).

Conflict of Interest

The authors declare that they have no conflicts of interest to disclose.

References

[1] Azargohar, R., Nanda, S., Kang, K., Bond, T., Karunakaran, C., Dalai, A.K., Kozinski, J.A., 2019. Effects of bio-additives on the physicochemical properties and mechanical behavior of canola hull fuel pellets. Renewable Energy. 132, 296-307.

[2] Bals, B.D., Gunawan, C., Moore, J., Teymouri, F., Dale, B.E., 2014. Enzymatic hydrolysis of pelletized AFEX^{TM}-treated corn stover at high solid loadings. Biotechnol. Bioeng. 111(2), 264-271.

[3] Baral, N.R., Chen, X., Palasz, J.M., Singh, R., Krishnamoorthy, A., Pidatala, V.R., Lewis, T.S.A., Dou, C., Ding, L., Choudhary, H., Sun, N., Simmons, B.A., Scown, C.D., 2025. Cost of deconstruction depots for diversified, waste-based lignocellulosic sugars using distillable solvents. ACS Sustain. Chem. Eng. 13(2), 13100-13111.

[4] Binder, J.B., Raines, R.T., 2010. Fermentable sugars by chemical hydrolysis of biomass. Proc. Natl. Acad. Sci. USA. 107(10), 4516-4521.

[5] Bozell, J.J., Petersen, G.R., 2010. Technology development for the production of biobased products from biorefinery carbohydrates-the US department of energy's "Top 10" revisited. Green Chem. 12(4), 539.

[6] Brandt, A., Gräsvik, J., Hallett, J.P., Welton, T., 2013. Deconstruction of lignocellulosic biomass with ionic liquids. Green Chem. 15(3), 550-583.

[7] Chandra, R.P., Esteghlalian, A.R., Saddler, J.N., 2008. Assessing substrate accessibility to enzymatic hydrolysis by cellulases, in: Hu, T.Q. (Ed.), Characterization of Lignocellulosic Materials. Wiley, pp. 60-80.

[8] Chen, W.H., Nižetić, S., Sirohi, R., Huang, Z., Luque, R., M Papadopoulos, A., Sakthivel, R., Phuong Nguyen, X., Tuan Hoang, A., 2022a. Liquid hot water as sustainable biomass pretreatment technique for bioenergy production: a review. Bioresour. Technol. 344, 126207.

[9] Chen, X. He, D., Hou, T., Lu, M., Mosier, N.S., Han, L., Xiao, W., 2022b. Structure-property-degradability relationships of varisized lignocellulosic biomass induced by ball milling on enzymatic hydrolysis and alcoholysis. Biotechnol. Biofuels Bioprod. 15, 36.

[10] Chen, X., Liu, S., Zhai, R., Yuan, X., Yu, Y., Shen, G., Wang, Z., Yu, J., Jin, M., 2022c. Lime pretreatment of pelleted corn stover boosts ethanol titers and yields without water washing or detoxifying pretreated biomass. Renewable Energy. 192, 396-404.

[11] Chen, X., Freitas Dos Santos, A.C., Ramirez Gutierrez, D.M., Song, P., Aston, J.E., Thompson, D.N., Dooley, J.H., Ladisch, M.R., Mosier, N.S., 2023a. Effect of pelleting on the enzymatic digestibility of corn stover. Bioresour. Technol. 384, 129338.

[12] Chen, X., Freitas dos Santos, A.C., Ramirez Gutierrez, D.M., Zhang, S., Aston, J.E., Thompson, D.N., Ladisch, M.R., Mosier, N.S., 2023b. Understanding the influence of water-soluble compounds from unpretreated corn stover pellets on enzymatic hydrolysis of cellulose. ACS Sustain. Chem. Eng. 11(50), 17616-17624.

[13] Chen, X., Krishnamoorthy, A., Palasz, J., Pidatala, V.R., Lewis, T., Tian, Y., Barcelos, C., Zhou, X., Kang, X., Han, Y., Dou, C., Choudhary, H., Sun, N., Sundstrom, E., Eudes, A., Simmons, B., 2025. Distillable amine-based solvents for effective pretreatment of multiple biomass feedstocks. Biofuel Res. J. 12(3), 2487-2502.

[14] Chen, X., Kuhn, E., Jennings, E.W., Nelson, R., Tao, L., Zhang, M., Tucker, M.P., 2016. DMR (deacetylation and mechanical refining) processing of corn stover achieves high monomeric sugar concentrations (230 g L^{-1}) during enzymatic hydrolysis and high ethanol concentrations (>10% v/v) during fermentation without hydrolysate purification or concentration. Energy Environ. Sci. 9(4), 1237-1245.

[15] Chen, X., Mosier, N., Ladisch, M., 2024. Valorization of lignin from aqueous-based lignocellulosic biorefineries. Trends Biotechnol. 42(11), 1348-1362.

[16] Chen, X., Yuan, X., Chen, S., Yu, J., Zhai, R., Xu, Z., Jin, M., 2021. Densifying Lignocellulosic biomass with alkaline Chemicals (DLC) pretreatment unlocks highly fermentable sugars for bioethanol production from corn stover. Green Chem. 23(13), 4828-4839.

[17] Chen, X., Zhang, Y., Hou, T., Han, L., Xiao, W., 2018. Catalysis performance comparison of a Brønsted acid H_2SO_4 and a Lewis acid $Al_2(SO_4)_3$ in methyl levulinate production from biomass carbohydrates. J. Energy Chem. 27(2), 552-558.

[18] Chundawat, S.P.S., Beckham, G.T., Himmel, M.E., Dale, B.E., 2011. Deconstruction of lignocellulosic biomass to fuels and chemicals. Annu. Rev. Chem. Biomol. Eng. 2, 121-145.

[19] da Silva, A.S., Espinheira, R.P., Teixeira, R.S.S., de Souza, M.F., Ferreira-Leitão, V., Bon, E.P.S., 2020. Constraints and advances in high-solids enzymatic hydrolysis of lignocellulosic biomass: a critical review. Biotechnol. Biofuels Bioprod. 13, 58.

[20] Dien, B.S., Mitchell, R.B., Bowman, M.J., Jin, V.L., Quarterman, J., Schmer, M.R., Singh, V., Slininger, P.J., 2018. Bioconversion of pelletized big bluestem, switchgrass, and low-diversity grass mixtures into sugars and bioethanol. Front. Energy Res. 6.

[21] Ding, K., Lin, H., Chen, X., Jia, X., Shi, S., Guo, X., Zhou, L., Han, L., Xiao, W., 2025. Dry alkali-ball milling for high-solid conversion of biomass to sugar and ethanol: comparison with conventional ball milling. Fuel. 401, 135931.

[22] Ding, K., Liu, D., Chen, X., Zhang, H., Shi, S., Guo, X., Zhou, L., Han, L., Xiao, W., 2024. Scalable lignocellulosic biorefineries: technoeconomic review for efficient fermentable sugars production. Renew. Sust. Energy Rev. 202, 114692.

[23] du Pasquier, J., Paës, G., Perré, P., 2023. Principal factors affecting the yield of dilute acid pretreatment of lignocellulosic biomass: a critical review. Bioresour. Technol. 369, 128439.

[24] Duque, A., Manzanares, P., Ballesteros, M., 2017. Extrusion as a pretreatment for lignocellulosic biomass: fundamentals and applications. Renewable Energy. 114(PartB), 1427-1441.

[25] Edwards, W., 2020. Estimating a Value for Corn Stover.

[26] Esteghlalian, A.R., Bilodeau, M., Mansfield, S.D., Saddler, J.N., 2001. Do enzymatic hydrolyzability and Simons' stain reflect the changes in the accessibility of lignocellulosic substrates to cellulase enzymes?. Biotechnol. Prog. 17(6), 1049-1054.

[27] García-Maraver, A., Popov, V., Zamorano, M., 2011. A review of European standards for pellet quality. Renewable Energy. 36(12), 3537-3540.

[28] Gong, C., Bryant, N., Meng, X., Bhagia, S., Pu, Y., Xin, D., Bender Koch, C., Felby, C., Thygesen, L.G., Ragauskas, A., Thomsen, S.T., 2021a. Double bonus: surfactant-assisted biomass pelleting benefits both the pelleting process and subsequent enzymatic saccharification of the pretreated pellets. Green Chem. 23(2), 1050-1061.

[29] Gong, C., Thomsen, S.T., Meng, X., Pu, Y., Puig-Arnavat, M., Bryant, N., Bhagia, S., Felby, C., Ragauskas, A.J., Thygesen, L.G., 2021b. Effects of different pelleting technologies and parameters on pretreatment and enzymatic saccharification of lignocellulosic biomass. Renewable Energy. 179, 2147-2157.

[30] Gong, C., Meng, X., Thygesen, L.G., Sheng, K., Pu, Y., Wang, L., Ragauskas, A., Zhang, X., Thomsen, S.T., 2023. The significance of biomass densification in biological-based biorefineries: a critical review. Renew. Sust. Energy Rev. 183, 113520.

[31] Gong, C., Thomsen, S.T., Thygesen, L.G., Felby, C., 2019. Effects of preheating on briquetting and subsequent hydrothermal pretreatment for enzymatic saccharification of wheat straw. Biotechnol. Prog. 35(4), e2808.

[32] Guo, H., He, T., Lee, D.J., 2022. Contemporary proteomic research on lignocellulosic enzymes and enzymolysis: a review. Bioresour. Technol. 344(Part B), 126263.

[33] Guragain, Y.N., Wilson, J., Staggenborg, S., McKinney, L., Wang, D., Vadlani, P.V., 2013. Evaluation of pelleting as a pre-processing step for effective biomass deconstruction and fermentation. Biochem. Eng. J. 77, 198-207.

[34] He, D., Chen, X., Lu, M., Shi, S., Cao, L., Yu, H., Lin, H., Jia, X., Han, L., Xiao, W., 2023. High-solids saccharification and fermentation of ball-milled corn stover enabling high titer bioethanol production. Renewable Energy. 202, 336-346.

[35] Holm, J.K., Henriksen, U.B., Hustad, J.E., Sørensen, L.H., 2006. Toward an understanding of controlling parameters in softwood and hardwood pellets production. Energy Fuels. 20(6), 2686-2694.

[36] Holtzapple, M.T., Jun, J.H., Ashok, G., Patibandla, S.L., Dale, B.E., 1991. The ammonia freeze explosion (AFEX) process. Appl. Biochem. Biotechnol. 28-29, 59-74.

[37] Huber, G.W., Iborra, S., Corma, A., 2006. Synthesis of transportation fuels from biomass: chemistry, catalysts, and engineering. Chem. Rev. 106(9), 4044-4098.

[38] Jiang, X., Zhai, R., Li, H., Li, C., Deng, Q., Jin, M., 2023. Understanding acid hydrolysis of corn stover during densification pretreatment for quantitative predictions of enzymatic hydrolysis efficiency using modified pretreatment severity factor. Bioresour. Technol. 386, 129487.

[39] Jørgensen, H., Kristensen, J.B., Felby, C., 2007. Enzymatic conversion of lignocellulose into fermentable sugars: challenges and opportunities. Biofuels, Bioprod. Bioref. 1(2), 119-134.

[40] Kaliyan, N., Morey, R.V., 2010. Natural binders and solid bridge type binding mechanisms in briquettes and pellets made from corn stover and switchgrass. Bioresour. Technol. 101(3), 1082-1090.

[41] Kaliyan, N., Vance Morey, R., 2009. Factors affecting strength and durability of densified biomass products. Biomass Bioenergy. 33, 337-359.

[42] Kenney, K., Cafferty, K.G., Jacobson, J.J., Bonner, I.J., Gresham, G.L., Smith, W.A., Thompson, D.N., Thompson, V.S., Tumuluru, J.S., Yancey, N., 2013. Feedstock supply system design and economics for conversion of lignocellulosic biomass to hydrocarbon fuels: conversion pathway: biological conversion of sugars to hydrocarbons "the 2017 design case". Idaho National Laboratory (United States).

[43] Kim, T.H., Kim, J.S., Sunwoo, C., Lee, Y.Y., 2003. Pretreatment of corn stover by aqueous ammonia. Bioresour. Technol. 90(1), 39-47.

[44] Kim, Y., Hendrickson, R., Mosier, N.S., Ladisch, M.R., 2009. Liquid hot water pretreatment of cellulosic biomass. Methods Mol. Biol. 581, 93-102.

[45] Krishnamoorthy, A., R. Pidatala, V., Chen, X., M. Palasz, J., Han, Y., Lewis, T., Choudhary, H., Rodriguez, A., Gladden, J.M., Dou, C., Sun, N., Simmons, B.A., 2025. Comparison between two different approaches for the deconstruction of lignocellulosic feedstocks using alkanolamine-based solvents. RSC. Sustainability. 3(9), 3915-3924.

[46] Kumar, L., Tooyserkani, Z., Sokhansanj, S., Saddler, J.N., 2012. Does densification influence the steam pretreatment and enzymatic hydrolysis of softwoods to sugars?. Bioresour. Technol. 121, 190-198.

[47] Ladisch, M.R., Lin, K.W., Voloch, M., Tsao, G.T., 1983. Process considerations in the enzymatic hydrolysis of biomass. Enzyme Microb. Technol. 5(2), 82-102.

[48] Lamers, P., Roni, M.S., Tumuluru, J.S., Jacobson, J.J., Cafferty, K.G., Hansen, J.K., Kenney, K., Teymouri, F., Bals, B., 2015. Techno-economic analysis of decentralized biomass processing depots. Bioresour. Technol. 194, 205-213.

[49] Liu, H., Chen, X., Ji, G., Yu, H., Gao, C., Han, L., Xiao, W., 2019. Mechanochemical deconstruction of lignocellulosic cell wall polymers with ball-milling. Bioresour. Technol. 286, 121364.

[50] Liu, S., Yu, Y., Xu, Z., Chen, S., Shen, G., Yuan, X., Deng, Q., Shen, W., Yang, S., Zhang, C., Chen, X., Jin, M., 2022. Efficient corncob biorefinery for ethanol initiated by a novel pretreatment of densifying lignocellulosic biomass with sulfuric Acid. Fermentation. 8(661), 661.

[51] Liu, Y., Cruz-Morales, P., Zargar, A., Belcher, M.S., Pang, B., Englund, E., Dan, Q., Yin, K., Keasling, J.D., 2021. Biofuels for a sustainable future. Cell. 184(6), 1636-1647.

[52] Li, C., Kerner, P., Williams, C.L., Hoover, A., Ray, A.E., 2020. Characterization and localization of dynamic cell wall structure and inorganic species variability in harvested and stored corn stover fractions as functions of biological degradation. ACS Sustain. Chem. Eng. 8(18), 6924-6934.

[53] Li, Y., Li, X., Shen, F., Wang, Z., Yang, G., Lin, L., Zhang, Y., Zeng, Y., Deng, S., 2014. Responses of biomass briquetting and pelleting to water-involved pretreatments and subsequent enzymatic hydrolysis. Bioresour. Technol. 151, 54-62.

[54] Lynd, L.R., Beckham, G.T., Guss, A.M., Jayakody, L.N., Karp, E.M., Maranas, C., McCormick, R.L., Amador-Noguez, D., Bomble, Y.J., Davison, B.H., Foster, C., Himmel, M.E., Holwerda, E.K., Laser, M.S., Ng, C.Y., Olson, D.G., Román-Leshkov, Y., Trinh, C.T., Tuskan, G.A., Upadhayay, V., Wyman, C.E., 2022. Toward low-cost biological and hybrid biological/catalytic conversion of cellulosic biomass to fuels. Energy Environ. Sci. 15(3), 938-990.

[55] Lyu, Q., Chen, X., Li, W., Zhang, Y., Xiao, A., Chen, J., Han, L., Zhou, C., Xiao, W., 2022. A multi-product strategy for the fractionation of corn stover based on peracetic acid and maleic acid processing. J. Environ. Chem. Eng. 10, 108764.

[56] Lyu, Q., Chen, X., Zhang, Y., Yu, H., Han, L., Xiao, W., 2021. One-pot fractionation of corn stover with peracetic acid and maleic acid. Bioresour. Technol. 320(Part A), 124306.

[57] Mamun, S., Hansen, J.K., Roni, M.S., 2020. Supply, operational, and market risk reduction opportunities: managing risk at a cellulosic biorefinery. Renew. Sust. Energy Rev. 121, 109677.

[58] Manandhar, A., Shah, A., 2017. Life cycle assessment of feedstock supply systems for cellulosic biorefineries using corn stover transported in conventional bale and densified pellet formats. J. Clean. Prod. 166, 601-614.

[59] Mokomele, T., da Costa Sousa, L., Colbert, A., Dale, B.E., Görgens, J.F., Balan, V., 2022. Coupling AFEX and steam-exploded sugarcane residue pellets with a room temperature CIIII-activation step lowered enzyme dosage requirements for sugar conversion. Chem. Eng. J. 446(Part 2), 137117.

[60] Morais, A.R.C., Zhang, J., Dong, H., Otto, W.G., Mokomele, T., Hodge, D., Balan, V., Dale, B.E., Lukasik, R.M., da Costa Sousa, L., 2022. Development of an ammonia pretreatment that creates synergies between biorefineries and advanced biomass logistics models. Green Chem. 24(11), 4443-4462.

[61] Mosier, N., Wyman, C., Dale, B., Elander, R., Lee, Y.Y., Holtzapple, M., Ladisch, M., 2005. Features of promising technologies for pretreatment of lignocellulosic biomass. Bioresour. Technol. 96(6), 673-686.

[62] Mosier, N.S., 2013. Fundamentals of aqueous pretreatment of biomass, in: Wyman, C.E. (Ed.), Aqueous Pretreatment of Plant Biomass for Biological and Chemical Conversion to Fuels and Chemicals. Wiley, pp. 129-143.

[63] Mujtaba, M., Fernandes Fraceto, L., Fazeli, M., Mukherjee, S., Savassa, S.M., Araujo de Medeiros, G., do Espírito Santo Pereira, A., Mancini, S.D., Lipponen, J., Vilaplana, F., 2023. Lignocellulosic biomass from agricultural waste to the circular economy: *a review with focus on biofuels, biocomposites and bioplastics*. J. Clean. Prod. 402, 136815.

[64] Nahar, N., Pandey, R., Pourhashem, G., Ripplinger, D., Pryor, S.W., 2021. Life Cycle Perspectives of Using Non-Pelleted vs. Pelleted Corn Stover in a Cellulosic Biorefinery. Energies. 14(9), 2518.

[65] Nahar, N., Pryor, S.W., 2017. Effects of reduced severity ammonia pretreatment on pelleted corn stover. Ind. Crops Prod. 109, 163-172.

[66] Nahar, N., Pryor, S.W., 2014. Reduced pretreatment severity and enzyme loading enabled through switchgrass pelleting. Biomass Bioenergy. 67, 46-52.

[67] Pandey, R., Nahar, N., Pryor, S., Pourhashem, G., 2021. Cost and environmental benefits of using pelleted corn stover for bioethanol production. Energies. 14(9), 2528.

[68] Pandey, R., Nahar, N., Tumuluru, J.S., Pryor, S.W., 2019. Quantifying reductions in soaking in aqueous ammonia pretreatment severity and

enzymatic hydrolysis conditions for corn stover pellets. Bioresour. Technol. Rep. 7, 100187.

[69] Paul, A., Juneja, A., Tumuluru, J.S., Kumar, D., 2023. A comprehensive evaluation of the various routes of biochemical conversion of pelletized biomass, in: Densification Impact on Raw, Chemically and Thermally Pretreated Biomass: Physical Properties and Biofuels Production, Sustainable Chemistry Series. WORLD SCIENTIFIC (EUROPE), pp. 291-316.

[70] Payne, C.M., Knott, B.C., Mayes, H.B., Hansson, H., Himmel, M.E., Sandgren, M., Ståhlberg, J., Beckham, G.T., 2015. Fungal cellulases. Chem. Rev. 115(3), 1308-1448.

[71] Petridis, L., Smith, J.C., 2018. Molecular-level driving forces in lignocellulosic biomass deconstruction for bioenergy. Nat. Rev. Chem. 2, 382-389.

[72] Pu, Y., Hu, F., Huang, F., Davison, B.H., Ragauskas, A.J., 2013. Assessing the molecular structure basis for biomass recalcitrance during dilute acid and hydrothermal pretreatments. Biotechnol. Biofuels. Bioprod. 6, 15.

[73] Qu, T., Zhang, X., Gu, X., Han, L., Ji, G., Chen, X., Xiao, W., 2017. Ball Milling for Biomass Fractionation and Pretreatment with Aqueous Hydroxide Solutions. ACS Sustain. Chem. Eng. 5(9), 7733-7742.

[74] Ragauskas, A.J., Williams, C.K., Davison, B.H., Britovsek, G., Cairney, J., Eckert, C.A., Frederick, W.J., Hallett, J.P., Leak, D.J., Liotta, C.L., Mielenz, J.R., Murphy, R., Templer, R., Tschaplinski, T., 2006. The path forward for biofuels and biomaterials. Science. 311(5760), 484-489.

[75] Ray, A.E., Hoover, A.N., Nagle, N., Chen, X., Gresham, G.L., 2013. Effect of pelleting on the recalcitrance and bioconversion of dilute-acid pretreated corn stover under low- and high-solids conditions. Biofuels. 4(3), 271-284.

[76] Rijal, B., Igathinathane, C., Karki, B., Yu, M., Pryor, S.W., 2012. Combined effect of pelleting and pretreatment on enzymatic hydrolysis of switchgrass. Bioresour. Technol. 116, 36-41.

[77] Rubin, E.M., 2008. Genomics of cellulosic biofuels. Nature. 454, 841-845.

[78] Ruiz, H.A., Galbe, M., Garrote, G., Ramirez-Gutierrez, D.M., Ximenes, E., Sun, S.N., Lachos-Perez, D., Rodríguez-Jasso, R.M., Sun, R.C., Yang, B., Ladisch, M.R., 2021. Severity factor kinetic model as a strategic parameter of hydrothermal processing (steam explosion and liquid hot water) for biomass fractionation under biorefinery concept. Bioresour. Technol. 342, 125961.

[79] Sarker, T.R., Azargohar, R., Dalai, A.K., Meda, V., 2021. Characteristics of torrefied fuel pellets obtained from co-pelletization of agriculture residues with pyrolysis oil. Biomass Bioenergy. 150, 106139.

[80] Scheller, H.V., Ulvskov, P., 2010. Hemicelluloses. Annu. Rev. Plant Biol. 61, 263-289.

[81] Scown, C.D., 2022. Prospects for carbon-negative biomanufacturing. Trends Biotechnol. 40(12), 1415-1424.

[82] Serra, L.A., da Silva Cruz, R.G., Gutierrez, D.M.R., Cruz, A.J.G., Canizares, C.A.T., Chen, X., Mosier, N., Thompson, D., Aston, J., Dooley, J., Sharma, P., De Marco, J.L., de Almeida, J.R.M., Erk, K., Ximenes, E., Ladisch, M.R., 2022. Screening method for Enzyme-based liquefaction of corn stover pellets at high solids. Bioresour. Technol. 363, 127999.

[83] Shahrukh, H., Oyedun, A.O., Kumar, A., Ghiasi, B., Kumar, L., Sokhansanj, S., 2016. Techno-economic assessment of pellets produced from steam pretreated biomass feedstock. Biomass Bioenergy. 87, 131-143.

[84] Sha, Y., Zhang, C., Xu, Z., Zhai, R., Jin, M., 2024. Quantitative assessment of ash effects on densifying lignocellulosic biomass with chemicals followed by autoclave (DLCA) pretreatment. Ind. Crops Prod. 216, 118767.

[85] Shen, G., Yuan, X., Cheng, Y., Chen, S., Xu, Z., Jin, M., 2023. Densification pretreatment with a limited deep eutectic solvent triggers high-efficiency fractionation and valorization of lignocellulose. Green Chem. 25(20), 8026-8039.

[86] Shen, G., Yuan, X., Chen, S., Liu, S., Jin, M., 2022. High titer cellulosic ethanol production from sugarcane bagasse via DLCA pretreatment and process development without washing/detoxifying pretreated biomass. Renewable Energy. 186, 904-913.

[87] Shi, J., Thompson, V.S., Yancey, N.A., Stavila, V., Simmons, B.A., Singh, S., 2013. Impact of mixed feedstocks and feedstock densification on ionic liquid pretreatment efficiency. Biofuels. 4(1), 63-72.

[88] Sokhansanj, S., Mani, S., Bi, X., Zaini, P., Tabil, L., 2005. Binderless pelletization of biomass., in: 2005 Tampa, FL July 17-20, 2005. Presented at the 2005 Tampa, FL July 17-20, 2005, American Society of Agricultural and Biological Engineers, St. Joseph, MI.

[89] Sundaram, V., Muthukumarappan, K., 2016a. Impact of AFEX™ pretreatment and extrusion pelleting on pellet physical properties and sugar recovery from corn stover, prairie cord grass, and switchgrass. Appl. Biochem. Biotechnol. 179, 202-219.

[90] Sundaram, V., Muthukumarappan, K., 2016b. Influence of AFEX™ pretreated corn stover and switchgrass blending on the compaction characteristics and sugar yields of the pellets. Ind. Crops Prod. 83, 537-544.

[91] Takada, M., Chandra, R., Wu, J., Saddler, J.N., 2020. The influence of lignin on the effectiveness of using a chemithermomechanical pulping based process to pretreat softwood chips and pellets prior to enzymatic hydrolysis. Bioresour. Technol. 302, 122895.

[92] Takada, M., Saddler, J.N., 2021. The influence of pre-steaming and lignin distribution on wood pellet robustness and ease of subsequent enzyme-mediated cellulose hydrolysis. Sustainable Energy Fuels. 5(2), 424-429.

[93] Tang, Y., Chandra, R.P., Sokhansanj, S., Saddler, J.N., 2018. Influence of steam explosion processes on the durability and enzymatic digestibility of wood pellets. Fuel. 211, 87-94.

[94] Theerarattananoon, K., Xu, F., Wilson, J., Ballard, R., Mckinney, L., Staggenborg, S., Vadlani, P., Pei, Z.J., Wang, D., 2011. Physical properties of pellets made from sorghum stalk, corn stover, wheat straw, and big bluestem. Ind. Crops Prod. 33(2), 325-332.

[95] Theerarattananoon, K., Xu, F., Wilson, J., Staggenborg, S., Mckinney, L., Vadlani, P., Pei, Z., Wang, D., 2012. Effects of the pelleting conditions on chemical composition and sugar yield of corn stover, big bluestem, wheat straw, and sorghum stalk pellets. Bioprocess Biosyst. Eng. 35, 615-623.

[96] Tumuluru, J.S., Conner, C.C., Hoover, A.N., 2016. Method to produce durable pellets at lower energy consumption using high moisture corn stover and a corn starch binder in a flat die pellet mill. J. Visualized Exp. (112), 54092.

[97] Tumuluru, J.S., Wright, C.T., Hess, J.R., Kenney, K.L., 2011. A review of biomass densification systems to develop uniform feedstock commodities for bioenergy application. Biofuels, Bioprod. Biorefin. 5(6), 683-707.

[98] Tumuluru, J.S., Yancey, N., 2018. Conventional and advanced mechanical preprocessing methods for biomass: performance quality attributes and cost analysis, in: Tumuluru, J.S. (Ed.), Biomass Preprocessing and Pretreatments for Production of Biofuels: Mechanical, Chemical and Thermal Methods. CRC Press, Boca Raton, FL : CRC Press, Taylor & Francis Group, "A science publishers book.," pp. 15-49.

[99] Tumuluru, J.S., 2023. Densification Impact on Raw, Chemically and Thermally Pre-treated Biomass: Physical Properties and Biofuels Production, Sustainable Chemistry Series. WORLD SCIENTIFIC (EUROPE).

[100] Tumuluru, J.S., 2020a. Biochemical and thermochemical conversion performance of densified products for biofuels production, in: Biomass Densification: Systems, Particle Binding, Process Conditions, Quality Attributes, Conversion Performance, and International Standards. Springer International Publishing, Cham, pp. 123-160.

[101] Tumuluru, J.S., 2020b. Binding mechanism, densification systems, process variables, and quality attributes, in: Biomass Densification: Systems, Particle Binding, Process Conditions, Quality Attributes, Conversion Performance, and International Standards. Springer International Publishing, Cham, pp. 23-61.

[102] Tumuluru, J.S., 2020c. Advances in biomass densification and its impact on cost and quality, in: Biomass Densification: Systems, Particle Binding, Process Conditions, Quality Attributes, Conversion Performance, and International Standards. Springer International Publishing, Cham, pp. 85-121.

[103] Tumuluru, J.S., 2014. Effect of process variables on the density and durability of the pellets made from high moisture corn stover. Biosyst. Eng. 119, 44-57.

[104] U.S. Department of Energy, 2020.

[105] Ullah, A., Zhang, Y., Liu, C., Qiao, Q., Shao, Q., Shi, J., 2023. Process intensification strategies for green solvent mediated biomass pretreatment. Bioresour. Technol. 369, 128394.

[106] Verdía Barbará, P., Choudhary, H., Nakasu, P.S., Al-Ghatta, A., Han, Y., Hopson, C., Aravena, R.I., Mishra, D.K., Ovejero-Pérez, A., Simmons, B.A., Hallett, J.P., 2025. Recent advances in the use of ionic liquids and deep eutectic solvents for lignocellulosic biorefineries and biobased chemical and material production. Chem. Rev. 125(12), 5461-5583.

[107] Wang, Z., Xu, Z., Chen, S., Chen, X., Yuan, X., Shen, G., Jiang, X., Liu, S., Jin, M., 2022. Effects of storage temperature and time on enzymatic digestibility and fermentability of Densifying lignocellulosic biomass with chemicals pretreated corn stover. Bioresour. Technol. 347, 126359.

[108] Wolfaardt, F.J., Leite Fernandes, L.G., Cangussu Oliveira, S.K., Duret, X., Görgens, J.F., Lavoie, J.-M., 2021. Recovery approaches for sulfuric acid from the concentrated acid hydrolysis of lignocellulosic feedstocks: a mini-review. Energy Convers. Manage. X. 10, 100074.

[109] Wolfrum, E.J., Nagle, N.J., Ness, R.M., Peterson, D.J., Ray, A.E., Stevens, D.M., 2017. The effect of biomass densification on structural sugar release and yield in biofuel feedstock and feedstock blends. BioEnergy Res. 10, 478-487.

[110] Wu, J., Chandra, R., Takada, M., Del Rio, P., Kim, K.H., Kim, C.S., Liu, L.-Y., Renneckar, S., Saddler, J., 2020. Alkaline sulfonation and thermomechanical pulping pretreatment of softwood chips and pellets to enhance enzymatic hydrolysis. Bioresour. Technol. 315, 123789.

[111] Wu, J., Ebadian, M., Kim, K.H., Kim, C.S., Saddler, J., 2022. The use of steam pretreatment to enhance pellet durability and the enzyme-mediated hydrolysis of pellets to fermentable sugars. Bioresour. Technol. 347, 126731.

[112] Xu, F., Sun, J., Konda, N.V.S.N.M., Shi, J., Dutta, T., Scown, C.D., Simmons, B.A., Singh, S., 2016. Transforming biomass conversion with ionic liquids: process intensification and the development of a high-gravity, one-pot process for the production of cellulosic ethanol. Energy Environ. Sci. 9, 1042-1049.

[113] Yancey, N.A., Tumuluru, J.S., Wright, C.T., 2013. Drying, grinding and pelletization studies on raw and formulated biomass feedstock's for bioenergy applications. J. Biobased Mat. Bioenergy. 7, 549-558.

[114] Yan, L., Ma, R., Li, L., Fu, J., 2016. Hot water pretreatment of lignocellulosic biomass: an effective and environmentally friendly approach to enhance biofuel production. Chem. Eng. Technol. 39(10), 1759-1770.

[115] Yuan, X., Chen, X., Shen, G., Chen, S., Yu, J., Zhai, R., Xu, Z., Jin, M., 2022. Densifying lignocellulosic biomass with sulfuric acid provides a durable feedstock with high digestibility and high fermentability for cellulosic ethanol production. Renewable Energy. 182, 377-389.

[116] Yuan, X., Shen, G., Chen, S., Shen, W., Chen, X., Liu, S., Jin, M., 2023. Elucidating the mechanism of densifying lignocellulosic biomass with acidic chemicals (DLC) for lignocellulosic biorefinery. Green Chem. 25(17), 6759-6773.

[117] Yuan, X., Shen, G., Huo, J., Chen, S., Shen, W., Zhang, C., Jin, M., 2024. Enhanced biomass densification pretreatment using binary chemicals for efficient lignocellulosic valorization. J. Bioresour. Bioprod. 9(4), 548-564.

[118] Yu, J., Chen, S., Yu, Y., Zhang, C., Jin, M., 2024. Influence of feedstock selection on cellulosic ethanol production based on densified biomass with calcium hydroxide and regular steam pretreatment. Renewable Energy. 227, 120561.

[119] Yu, J., Xu, Z., Chen, S., Yu, Y., Zhang, C., Chen, X., Jin, M., 2022a. DLCA (ch) pretreatment brings economic benefits to both biomass logistics and biomass conversion for low cost cellulosic ethanol production. Fuel. 311, 122603.

[120] Yu, Y., Wu, J., Ren, X., Lau, A., Rezaei, H., Takada, M., Bi, X., Sokhansanj, S., 2022b. Steam explosion of lignocellulosic biomass for multiple advanced bioenergy processes: A review. Renew. Sust. Energy Rev. 154, 111871.

[121] Zeng, M., Pan, X., 2022. Insights into solid acid catalysts for efficient cellulose hydrolysis to glucose: progress, challenges, and future opportunities. Catal. Rev. 64(3), 445-490.

[122] Zhai, R., Hu, J., Jin, M., 2022. Towards efficient enzymatic saccharification of pretreated lignocellulose: Enzyme inhibition by lignin-derived phenolics and recent trends in mitigation strategies. Biotechnol. Adv. 61, 108044.

[123] Zhang, J., Mohammadi, M., Gong, H., Hodge, D.B., Tumuluru, J., da Costa Sousa, L., Dale, B., Balan, V., 2025. High throughput pretreatment of corn stover using compacted biomass with recycled ammonia (COBRA) process. Chem. Eng. J. 505, 159731.

[124] Zhang, S., Sheng, K., Chen, X., Zhang, X., Mosier, N.S., 2021. Conversion of glucose to 5-hydroxymethyl furfural in water-acetonitrile-dimethyl sulfoxide solvent with aluminum on activated carbon and maleic acid. Ind. Crops Prod. 174, 114220.

[125] Zhong, C., Lau, M.W., Balan, V., Dale, B.E., Yuan, Y.J., 2009. Optimization of enzymatic hydrolysis and ethanol fermentation from AFEX-treated rice straw. Appl. Microbiol. Biotechnol. 84, 667-676.

[126] Zhou, Z., Liu, D., Zhao, X., 2021. Conversion of lignocellulose to biofuels and chemicals via sugar platform: An updated review on chemistry and mechanisms of acid hydrolysis of lignocellulose. Renew. Sust. Energy Rev. 146, 111169.

[127] Zhu, J.Y., Pan, X., 2022. Efficient sugar production from plant biomass: current status, challenges, and future directions. Renew. Sust. Energy Rev. 164, 112583.

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