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A Review of Bioethanol Derived from Lignocellulosic Agricultural Waste: Fermentation Pathways, Microbial Roles, Bioreactor Design, Process Integration, and Industrial Implementation Challenge

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Abstract

Growing concerns over fossil fuel depletion and environmental sustainability have accelerated the development of lignocellulosic bioethanol as a renewable energy alternative. This review critically evaluates recent advances in lignocellulosic bioethanol production, highlighting key technological developments, industrial challenges, and future research directions toward sustainable commercialization. This review synthesizes evidence from 152 references, of which 86.7% were published within the last 10 years. Among the evaluated feedstocks, cereal crop residues (rice, corn, wheat, barley, oat, sorghum) and sugar crops (sugarcane and oil palm) generally exhibited superior bioethanol performance, achieving ethanol yields of 0.28–0.57 g/g, concentrations of 23.1–93.0 g/L, and fermentation efficiencies of 21.5–89.6%, whereas fibrous residues (cotton, kenaf, sunflower, ramie, sesame, groundnut) typically produced lower ethanol concentrations ranging from 1.9–63.0 g/L. Conventional and advanced bioreactor configurations provide complementary advantages for improving mass transfer, fermentation stability, and scale-up performance in lignocellulosic bioethanol production. Furthermore, integrating hydrolysis and fermentation with automated process control and AI-assisted optimization enhances ethanol productivity, process efficiency, and industrial applicability. Overall, the industrial implementation of lignocellulosic bioethanol is governed not only by ethanol yield but also by the ability to simultaneously optimize techno-economic performance, environmental sustainability, and process integration. Although recent studies have demonstrated substantial improvements through advanced pretreatment, optimized enzyme utilization, and process intensification; commercialization remains constrained by high enzyme and pretreatment costs, process complexity, feedstock variability, and the lack of standardized sustainability assessment methodologies across different production systems.

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Introduction

The replacement of fossil fuels is driven by growing interest in environmentally friendly and sustainable fuels. Renewable and sustainable energy sources are expected to form the foundation of future global bioenergy systems, one of which is bioethanol, produced from natural biomass based materials such as lignocellulose. Bioethanol has a higher oxygen content, leading to cleaner combustion and reduced emissions of pollutants such as carbon monoxide (CO) and nitrogen oxides (NOx) (Humera Farheen et al., 2026). Lignocellulose based raw materials are among the most promising bioenergy alternatives due to their abundance in nature, their classification as non-food resources, and their ability to minimize competition with food supplies (Maheshwari, 2018).

The bioethanol production process goes through two stages, namely upstream and downstream. The upstream stage involves biomass degradation, whereas the downstream stage involves fermentation. Biomass degradation often generates complex sugars and inhibitory compounds, which represent major challenges for the fermentation process (Humera Farheen et al., 2026; Iriel et al., 2024). Fermentation, as the downstream stage, is crucial to the success of bioethanol production because it enables the conversion of hydrolyzed sugars into ethanol (Maheshwari, 2018). Therefore, the development of fermentation methods that can streamline the bioethanol production process is necessary, including approaches such as co-fermentation and inhibitor-tolerant systems. With effective studies focusing on the operational aspects of fermentation, bioethanol production has strong potential for implementation at an industrial scale (Kassim et al., 2021; Pino et al., 2018). Bioethanol production begins with feedstock processing, where pretreatment and hydrolysis are used to break down lignocellulosic biomass into simple sugars. These sugars are then fermented by microorganisms to produce ethanol. After fermentation, the ethanol is purified and recovered to reach the required purity for industrial use. Sustainability in bioethanol production is determined not only by the fermentation system as the central process but also by the involvement of biological sources and the technological structure of the processes employed. Ethanol productivity and operational stability are largely determined by several factors, including fermentation pathways, the roles of biological sources such as enzymes and microorganisms, comparative bioethanol yield, bioreactor design, and process integration. This article further examines the future challenges and opportunities in lignocellulosic bioethanol fermentation systems.

This review provides a comprehensive overview of bioethanol production from lignocellulosic agricultural waste. It comprehensively examines feedstock characteristics, microbial agents, fermentation pathways, bioreactor designs, and process integration strategies. Unlike previous reviews that primarily focused on individual aspects such as feedstocks, pretreatment technologies, or fermentation processes, this review integrates these key components to provide a holistic evaluation of production efficiency, sustainability, and the future prospects of lignocellulosic bioethanol production. Particular emphasis is placed on the role of process integration in improving production efficiency, product consistency, and industrial-scale process stability. In addition, this review identifies current research gaps and highlights future research priorities and policy directions, providing a valuable scientific reference for advancing sustainable bioethanol production and accelerating the transition toward a circular bioeconomy.

Methods

This review was conducted using a literature-based approach to comprehensively evaluate the development of lignocellulosic bioethanol production from agricultural residues. Relevant publications were collected from various scientific databases and publishers, including ScienceDirect, Springer, Wiley, MDPI, Taylor & Francis, Google Scholar, and other peer-reviewed scientific sources. The selected literature focused on studies related to feedstock characteristics, pretreatment methods, hydrolysis processes, fermentation strategies, microorganisms, process optimization, and recent technological developments associated with lignocellulosic bioethanol production.

The literature search employed combinations of the following keywords using Boolean operators ("AND" and "OR") such as "bioethanol", "lignocellulosic biomass", "agricultural waste", "agricultural residues", "pretreatment", "enzymatic hydrolysis", "fermentation", "simultaneous saccharification and fermentation (SSF)", "separate hydrolysis and fermentation (SHF)", "consolidated bioprocessing (CBP)", "microorganisms", "yeast", "bacteria", "bioreactor", "process integration", "biofuel", and "renewable energy". A total of 152 references were included in this review and critically analyzed to identify major trends and technological progress in the field. To ensure adequate coverage of recent developments, 124 references (86.7%) were published during 2017–2026, while 65 references were published between 2022–2026. This distribution indicates that the review incorporates both foundational studies and recent advances, providing a comprehensive perspective on sustainable bioethanol production from lignocellulosic

agricultural residues. Earlier landmark publications published before 2016 were also included when they introduced fundamental concepts, established widely accepted methodologies, or provided historical perspectives essential for understanding the development of lignocellulosic bioethanol technologies.

The inclusion criteria consisted of (i) peer-reviewed journal articles published in English; (ii) studies focusing on lignocellulosic agricultural residues as bioethanol feedstocks; (iii) research investigating pretreatment technologies, hydrolysis, fermentation pathways, microbial applications, bioreactor systems, process integration, scale-up challenges, and operational constraints; and (iv) articles providing experimental results, comparative analyses, or comprehensive review discussions. Conversely, the exclusion criteria included conference abstracts, editorials, dissertations, book chapters, duplicated studies, and articles lacking sufficient methodological information.

The literature selection process involved four sequential stages. First, publications were identified through database searches using predefined keywords. Second, duplicate records were removed. Third, titles and abstracts were screened to assess their relevance to the review objectives. Finally, full-text articles were critically evaluated based on the inclusion and exclusion criteria before being selected for detailed analysis. The selected literature was synthesized using a thematic qualitative synthesis approach. The publications were categorized into six major themes: (i) lignocellulosic agricultural feedstocks and biomass characteristics, (ii) pretreatment technologies, (iii) hydrolysis and fermentation pathways, (iv) microbial roles and metabolic performance, (v) bioreactor design and process integration strategies, and (vi) scale-up challenges and operational constraints. Comparative analyses were conducted to identify technological advances, current limitations, research gaps, and emerging opportunities for improving the efficiency and sustainability of lignocellulosic bioethanol production.

Results and Discussion

Fermentation Pathways for Bioethanol Production

Conventional Fermentation of Reducing Sugars

Bioethanol fermentation under anaerobic conditions proceeds optimally when appropriate temperature and pH parameters are maintained, specifically a temperature range of 30–50℃ and a pH of 4.5–9.5. Higher initial sugar concentrations (150 g/L) can also yield high ethanol production, but the optimal concentration may vary depending on the microbial strain and other conditions (Zentou et al., 2017). Ethanol formation via the Embden–Meyerhof–Parnas (EMP) pathway requires microbial activity like *Lactobacillus plantarum* and *Thermoanaerobacterium saccharolyticum*; therefore, several parameters must be considered to optimize ethanol productivity, including sugar concentration, inoculum size, and fermentation time (Cui et al., 2020). Ethanol production involves multiple process stages. Lignocellulosic biomass first undergoes hydrolysis during the upstream process, after hydrolyzed sugars enter microbial cells and undergo glycolysis to form the intermediate compound pyruvate ($C_3H_3O_3^-$) (Jeevitha et al., 2023; Palupi et al., 2023). Pyruvate is subsequently converted into acetaldehyde through decarboxylation and finally reduced to ethanol, a process that regenerates Nicotinamide Adenine Dinucleotide (NAD^+). The formation of NAD^+ strongly supports the sustainability of bioethanol production, however factors such as sugar concentration must also be considered, as excessive levels can increase osmotic pressure in microbial cells and thereby disrupt process stability (Y. Liu et al., 2024).

Although high initial sugar concentrations can increase ethanol titer, they do not always improve overall fermentation performance. Several studies reported that excessive sugar levels create osmotic stress, reduce microbial viability, and prolong fermentation, particularly in strains with limited osmotolerance. Therefore, the optimum sugar concentration depends not only on substrate availability but also on microbial physiology, inoculum density, and inhibitor levels present in hydrolysates (Camargos et al., 2021; Frohman & Mira de Orduña, 2013). Therefore, despite continuous improvements in fermentation optimization, conventional hexose fermentation remains the most mature and industrially reliable pathway because of its high ethanol productivity and process robustness. However, its inability to efficiently utilize pentose ($C_5H_{10}O_5$) sugars limits carbon conversion from lignocellulosic biomass, making alternative fermentation strategies increasingly important (Z. L. Liu et al., 2014).

Pentose Fermentation and Co-Fermentation Schemes

Pentose fermentation has disadvantages compared to glucose ($C_6H_{12}O_6$) fermentation, due to additional biochemical complexity, lower fermentation rates, and increased byproduct formation. Complex biochemistry occurs when xylose and arabinose entering the microbial cell are converted into intermediates of central carbon (C) metabolism. Xylose can also undergo reduction to xylitol ($C_5H_{12}O_5$) in yeast systems, which then becomes xylulose after oxidation, followed by phosphorylation and channeling into the pentose

phosphate pathway before entering glycolysis. The efficiency of enzymes involved in pentose phosphate pathways (PPP) such as *transaldolase*, *transketolase*, ribose-5-phosphate ketol-isomerase, and d-ribulose-5-phosphate 3-epimerase, can significantly affect the rate of xylose metabolism. Overexpression of these enzymes has been shown to improve xylose consumption rates, but the rates are still generally lower than those for glucose (Bae et al., 2021; Kobayashi et al., 2018). Compared with glucose fermentation, pentose fermentation suffers from lower adenosine triphosphate (ATP) generation efficiency, slower sugar uptake, and more complex metabolic regulation. Consequently, although metabolic engineering has substantially improved xylose utilization, ethanol productivity generally remains below that of conventional glucose fermentation. This indicates that improving pentose metabolism requires coordinated optimization of transport systems, cofactor balance, and downstream carbon flux rather than single-gene modifications alone (Verho et al., 2003; Zhou & Lü, 2021). This also explains why improvements reported in individual studies are often difficult to reproduce across different microbial strains and lignocellulosic hydrolysates, where variations in sugar composition, inhibitor profiles, and metabolic regulation substantially influence fermentation performance.

Process integration methods aimed at reducing process complexity are designed to improve overall carbon efficiency. Consolidated Bio-Processing (CBP) integrates enzyme production, hydrolysis, and fermentation into a single process within a single reactor. Fermentation integration is also achieved through co-fermentation, which enables the simultaneous utilization of hexose and pentose sugar mixtures (Caro et al., 2024). CBP minimizes process units and enzyme costs but currently suffers from relatively low ethanol productivity due to limited availability of microorganisms capable of simultaneously producing cellulases and ethanol efficiently (Papathoti et al., 2024).

In Simultaneous Saccharification and Co-Fermentation (SSCF), enzymatic hydrolysis and fermentation occur concurrently in the same reactor. Performing enzymatic hydrolysis and fermentation simultaneously reduces the inhibition of enzymes by the sugars produced during hydrolysis, leading to more efficient conversion (Afedzi & Parakulsuksatid, 2023). Precise control is essential in SSCF with respect to both environmental and process conditions, such as temperature and pH. Effective environmental control is expected to minimize preferential glucose consumption caused by carbon catabolite repression. Meanwhile, maintaining optimal temperature and pH conditions enhances enzyme activity and microbial metabolism. Typically, temperatures are kept below 36℃ to balance the needs of both processes. Additional challenges must also be addressed during SSCF to optimize process performance and scalability, as High solid loading can improve ethanol concentration but poses mixing and viscosity challenges. Fed batch modes and enzyme feeding strategies help manage these issues (Jia et al., 2025). Despite its higher carbon efficiency, SSCF introduces operational compromises because cellulolytic enzymes generally perform best at 45–50℃, whereas most ethanologenic microorganisms grow optimally near 30–35℃. Consequently, SSCF often sacrifices either enzymatic hydrolysis efficiency or microbial fermentation performance. Future development therefore focuses on thermotolerant microorganisms and engineered enzymes capable of operating within overlapping optimal temperature ranges (Qiang et al., 2025). Therefore, the overall advantage of SSCF depends on whether the benefits of reduced end-product inhibition outweigh the compromises associated with non-optimal operating conditions for either enzymes or fermentative microorganisms.

Inhibitory Compounds and Fermentation Resilience

Inhibitor compounds represent the greatest obstacle to an efficient fermentation process. These inhibitors, including furan derivatives, weak organic acids, and phenolic compounds are generated during the decomposition of biomass components such as carbohydrates and lignin. Their presence negatively affects fermentation performance. Furan derivatives like furfural and 5-hydroxymethl-2-furaldehyde (HMF) are toxic to fermentative microorganisms, leading to decreased bioethanol production. Furfural is particularly potent, with high concentrations causing complete inhibition of ethanol fermentation. Furan derivatives inhibit microbial growth and delay sugar consumption, impacting overall fermentation efficiency (Z. Liu et al., 2021). Weak organic acids such as acetic acid (CH_3COOH), formic acid (HCOOH), and levulinic acid ($C_5H_8O_3$) increase intracellular acidity by lowering the pH and delay bioethanol productivity, thereby elevating cellular energy requirements for maintenance (H. Huang et al., 2011; L. Q. Wang et al., 2020). Phenolic compounds can damage microbial cell membranes and inhibit enzymatic activity. Vanillin, syringaldehyde, and phenol was highly inhibitory. For instance, o.1 g/L of catechin and 0.5 g/L of gallic acid inhibit fermentation (Bellido et al., 2018; Gou et al., 2015). The inhibitory effects reported across different studies vary considerably because inhibitor toxicity depends not only on compound concentration but also on inhibitor combinations, microbial strain tolerance, pretreatment method, and fermentation conditions. Hydrolysates rarely contain a single inhibitor; instead, multiple compounds often exert

synergistic effects that produce greater inhibition than individual compounds tested separately. Consequently, effective inhibitor management should focus on the overall inhibitor profile of lignocellulosic hydrolysates rather than targeting individual compounds in isolation (Straathof, 2023; L. Q. Wang et al., 2020).

The resilience of the fermentation process supporting sustainable bioethanol production is largely determined by operational parameters, adaptive evolution, and metabolic engineering. Key operational parameters that must be controlled include maintaining a pH of 4.5–9.5, temperature range of 30–50℃, and the gradual feeding of microorganisms (Zentou et al., 2017). Detoxification methods like 5% w/v activated carbon treatment significantly reduce the concentration of 77.9% furan derivatives and 98.6% aromatic monomers (Y. Zhang et al., 2018). Another research shows that enzymes like laccase or microorganisms such as *Trichoderma reesei* can effectively detoxify hydrolysates while simultaneously improving fermentation performance (Parawira & Tekere, 2011). Detoxification effectively improves fermentation performance but also introduces additional processing steps, chemical consumption, wastewater generation, and operational costs. Consequently, recent studies increasingly emphasize developing inhibitor-tolerant microorganisms rather than relying solely on extensive detoxification processes (J. Li et al., 2018). Strain adaptation represents one of the most promising strategies for mitigating inhibitory effects because evolving microbial strains to tolerate higher concentrations of inhibitors can substantially improve fermentation efficiency. Adapted strains show better performance in the presence of inhibitors. Similarly, the use of high-cell-density cultures or cell encapsulation can further alleviate inhibitor toxicity, enhance cell stability, and improve ethanol productivity under industrial fermentation conditions (Soares et al., 2020).

Biological Agents (Enzymes, Bacteria, Microorganisms) in Bioethanol Production
Enzymatic Contribution to Fermentation Efficiency

Enzymes function as biocatalysts that enhance reaction kinetics without being consumed during biochemical reactions. In bioethanol production, enzymes catalyze the hydrolysis of complex polysaccharides into reducing sugars that can be readily fermented by microorganisms. The rigid and recalcitrant structure of lignocellulosic biomass limits direct microbial fermentation and therefore necessitates enzymatic hydrolysis. Plant cell walls consist of a flexible primary layer composed mainly of cellulose and hemicellulose, and a thicker secondary layer reinforced with lignin (J. Li et al., 2023). Fig. 1 shows that lignin carbohydrate matrix provides strong structural resistance, requiring enzymatic hydrolysis to release fermentable sugars.

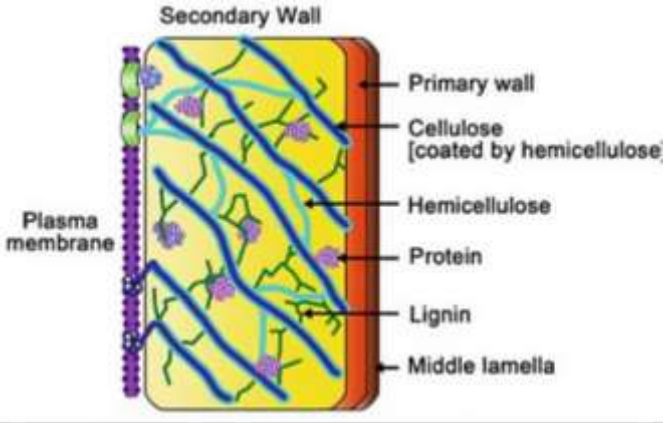

Figure 1. *Schematic Diagram of the Lignocellulosic Plant Cell Wall Structure* (*Biotechnology in China III: Biofuels and Bioenergy - Google Buku*, n.d.)

During the enzymatic hydrolysis of cellulose and hemicellulose, reducing sugars are produced. Reducing sugars includes all monosaccharides (glucose, fructose, galactose, mannose, ribose) and some disaccharides (lactose, maltose), whereas sucrose and starch are not reducing sugars. The availability of reducing sugars directly affects ethanol yield, fermentation kinetics, and overall process efficiency (Keerthi Patel M et al., 2025; Palupi et al., 2023). Hydrolysis process is influenced by several factors, including temperature, pH, enzyme loading, and resistance to inhibitors formed during pretreatment. For instance, an enzyme dose of 25 FPU/g glucan was optimal for *Eucalyptus grandis* hydrolysis (Bacquerié et al., 2025). Similarly, enzyme dosages ranging from 6.92–20 FPU/g of glucan were tested for wheat straw, with higher dosages leading to better yield (Coimbra et al., 2016). Increasing enzyme loading generally enhances sugar

release but exhibits diminishing returns beyond the optimum dosage because substrate accessibility rather than catalytic activity becomes the limiting factor. Consequently, excessive enzyme addition substantially increases operating costs without proportional improvements in ethanol yield. Therefore, optimizing enzyme dosage rather than maximizing enzyme addition is generally considered a more economically viable strategy for large-scale bioethanol production (Han et al., 2020).

As key intermediaries in fermentation, enzymes provide fermentable sugars in a continuous and controlled manner, thereby increasing ethanol yield without directly participating in the fermentation reaction. Efficient pretreatment strategies, including dilute acid, alkaline, and steam-based processes, are essential to alter the lignocellulosic structure by removing hemicellulose and reducing lignin content. These treatments enhance cellulose exposure by increasing porosity and decreasing structural recalcitrance, thereby facilitating enzymatic hydrolysis. For instance, alkaline pretreatment applied to Napier grass achieved higher lignin removal than untreated biomass, leading to improved hydrolysis performance (Bacquerié et al., 2025). Therefore, effective bioethanol production requires balancing pretreatment severity and enzymatic digestibility. Severe pretreatment improves cellulose accessibility but simultaneously promotes inhibitor formation, whereas mild pretreatment preserves sugar integrity but leaves residual lignin that limits enzyme accessibility.

Yeast-Based Fermentation Systems

The majority of industrial fermentation processes employ yeast as the primary biological agent. Yeast, especially *Saccharomyces cerevisiae*, is well known for its ability to convert sugars into ethanol and carbon dioxide (CO_2) during fermentation. This process is most effective under anaerobic conditions. Its capacity to enhance ethanol yield, optimize metabolic efficiency, and tolerate acidic conditions makes yeast a dominant microorganism in industrial fermentation (Satwika et al., 2019).

The high ethanol productivity of *Saccharomyces cerevisiae* makes it the most widely used yeast in bioethanol production for the fermentation of hexose sugars (Elhalis, 2024). In addition, non-conventional yeasts, such as *Scheffersomyces stipitis* and *Kluyveromyces marxianus*, have also attracted the attention of several researchers because of their high temperature resistance and ability to ferment pentose sugars (Nurcholis et al., 2020; Ochoa-Chacón et al., 2022). As a result, yeast engineering has become important to improve performance and maintain stable fermentation. An effective process achieves high ethanol yields, low inhibitor formation, and efficient use of mixed sugars. Under anaerobic conditions, yeast breaks down one molecule of glucose ($C_6H_{12}O_6$) into two molecules of ethanol (C_2H_5OH) and two molecules of carbon dioxide (CO_2), which represents the core biochemical reaction in bioethanol production as shown in Fig. 2.

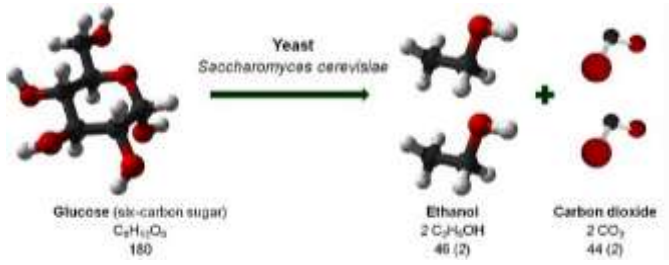

Figure 2. Conversion of Glucose into Ethanol and Carbon Dioxide by Yeast (*Bioenergy Research Group*, n.d.)

Although *Saccharomyces cerevisiae* remains the industrial standard because of its robustness and high ethanol productivity, its inability to naturally ferment pentose sugars limits complete utilization of lignocellulosic hydrolysates. In contrast, non-conventional yeasts can metabolize pentoses but generally exhibit lower ethanol productivity and weaker tolerance toward inhibitors, illustrating the trade-off between substrate versatility and industrial robustness (S. Sharma & Arora, 2020). Consequently, current research increasingly focuses on engineering *Saccharomyces cerevisiae* to combine its inherent robustness with enhanced pentose-fermenting capability, thereby improving the overall utilization of lignocellulosic hydrolysates.

Bacterial and Non-Conventional Fermentative Microorganisms

Bacteria can be a viable alternative in bioethanol fermentation systems, if they are capable of degrading a wide variety of sugars. This potential is supported by the diversity of metabolic pathways and the high biochemical flexibility of bacteria. These bacteria operate at higher temperatures, which can enhance the efficiency of bioethanol production (Kawada-Matsuo et al., 2017; Scully & Orlygsson, 2015). However, the main obstacle to implementation on an industrial scale is the low ethanol standards and greater

sensitivity to process parameters. Bacteria are also more sensitive to changes in environmental conditions such as pH, temperature, and substrate concentration, which can significantly affect fermentation performance (Dutta & Suresh Kumar, 2023; Scully & Orlygsson, 2015). Compared with yeast, bacterial ethanologens generally possess broader substrate utilization and faster metabolic rates. However, they often produce multiple fermentation byproducts, require stricter process control, and exhibit lower industrial stability. Consequently, bacterial systems are promising for integrated lignocellulosic biorefineries but currently remain less commercially established than yeast-based fermentation (W. C. Huang & Tang, 2006).

Bacteria and other microorganisms can significantly contribute to the development of SSCF strategies and bioethanol production technologies due to their metabolic flexibility and ease of genetic engineering . *Zymomonas mobilis* and recombinant *Escherichia coli* are among the most well-known bacteria involved in bioethanol fermentation. Recombinant strains of Z. mobilis have been developed to tolerate high temperatures and low nutritional conditions, which are common stress factors during fermentation (Kaewchana et al., 2021). Recombinant E. coli strains have been engineered to produce ethanol at near-theoretical yields, these strains are capable of co-fermenting hexoses and pentoses (Yanase, 2014). While *Clostridium* species such as *Clostridium acetobutylicum* and *Clostridium thermocellum,* have been extensively studied that their genetic modifications could enhanced ethanol, by improving biotin synthesis and other metabolic pathways (Chung et al., 2025). As a bacterium capable of producing large amounts of ethanol, *Zymomonas mobilis* utilizes the Entner–Doudoroff (ED) pathway, which is more thermodynamically favorable compared to EMP pathway (Jacobson et al., 2019). However, unlike recombinant E. coli, it requires additional genetic engineering to efficiently degrade complex sugars like xylose and mannose (Lou et al., 2025). *Clostridium* on the other hand has a disadvantage compared to conventional ethanol producing bacteria because it generates various byproducts such as acetate (CH_3COO^-), lactate ($C_3H_5O_3^-$), and hydrogen (H) which can reduce the overall yield of ethanol (Cheng et al., 2019). Some species, like C. thermocellum, can directly convert cellulose to ethanol through Consolidated Bioprocessing (CBP), which simplifies the process but still results in multiple byproducts (Papathoti et al., 2024). No single bacterial platform currently satisfies all industrial requirements. Z. *mobilis* provides superior ethanol productivity but limited substrate utilization, whereas recombinant E. *coli* ferments diverse sugars at the expense of greater metabolic complexity. Selection therefore depends on feedstock composition and process objectives rather than ethanol yield alone (Xia et al., 2019). These contrasting characteristics demonstrate that the selection of bacterial platforms should be based on specific feedstock characteristics and process configurations rather than a single performance indicator.

Strain Improvement and Adaptive Engineering Strategies

Microbial engineering is essential in the field of bioethanol fermentation. The primary objective of this approach is to increase bioethanol production by reducing inhibitory compounds. Adaptive Laboratory Evolution (ALE) is a systematic approach used to enhance microbial robustness by continuously exposing cells to stressful conditions, allowing naturally occurring adaptive traits to emerge over time. For example, strain 40B of *Saccharomyces cerevisiae* was developed through ALE and exhibited increased resistance to high inhibitor concentrations, resulting in improved cell growth and fermentation efficiency (Z. Zhang et al., 2026). Additionally, deletion of genes involved in proline or myo-inositol synthesis weakened strain tolerance against inhibitors, while their overexpression conferred increased tolerance (X. Wang et al., 2015). Adaptive laboratory evolution (ALE) provides stable improvements without requiring extensive genetic manipulation; however, the adaptive traits obtained are often strain-specific and may not be universally transferable to other industrial microorganisms. In contrast, metabolic engineering enables targeted pathway optimization but frequently requires complex regulatory balancing to maintain cellular fitness (Hossain, 2023; Padhan et al., 2025).

Application through a combination of genetic engineering and process engineering will result in higher ethanol yields, fewer byproducts, and increased overall conversion. Process engineering as a process improvement strategy, employs sustainable methodologies to modify operating conditions, inhibitor concentrations, co-cultivation strategies, and related factors (H. Dey et al., 2025). Future research is increasingly directed toward integrating strain engineering, process optimization, and reactor design rather than optimizing each component independently. Such integrated strategies are expected to maximize carbon conversion efficiency while simultaneously reducing enzyme consumption, inhibitor sensitivity, and overall production costs, thereby improving the commercial feasibility of lignocellulosic bioethanol production. Consequently, future commercial bioethanol production is likely to rely on integrated biological and process engineering strategies rather than improvements in individual technologies alone.

Comparative Bioethanol Outcomes from Agricultural Lignocellulosic Feedstocks

The use of different lignocellulosic feedstocks shows significant differences in many result parameters as shown in Table 1. These differences are influenced by internal feedstock characteristics such as chemical composition, structural complexity, and sugar availability. The varying proportions of sugars and lignin in each feedstock also determine the efficiency of enzymatic hydrolysis and microbial fermentation. This is evident in the high glucose content of some feedstock can yield high ethanol production under appropriate hydrolysis and fermentation conditions. Therefore, appropriate pretreatment and process engineering are necessary to achieve balanced performance (Dessie et al., 2024).

Table 1. Comparison of Feedstock Types, Operating Conditions, and Performance in Bioethanol Production from Lignocellulosic Agricultural Residues

Feedstock	Operating Conditions	Performance	Ref.
Rice straw	Pretreated using 4% NaOH alkali, soaked for 4 h, and autoclaved for 30 min. Hydrolyzed with (2:1:1) enzyme loading, 20% solid loading. SSF (Simultaneous Saccharification and Fermentation) by *S. cerevisiae* for 1 week at 30℃ and pH 4.5	A fermentation efficiency of 21.45% ethanol was attained	(Ningthoujam et al., 2023)
Rice straw	Pretreated by chemical (deep eutectic solvent), physical (microwave irradiation) and biological (laccase) pretreatment. Saccharified by cellulase/xylanase from *A. japonicus* and ethanol-fermented by *S. cerevisiae* and Pichia stipites at 48.36℃, and pH 6.03	Ethanol yield of 214 mg/g rice straw biomass (bioconversion efficiency 72.5%)	(Sawhney et al., 2023)
Rice straw	Pretreated with Choline chloride (ChCl): Acetic acid (AA)-based deep eutectic solvent under optimized conditions (ChCl:AA, Molar ratio of 1:3.59, 126℃, 2.5 h). Fermented by novel bacterial crude recombinant hydrolytic enzyme cocktail at 33.7℃ for 12.2 h	Ethanol yield of 182 L ethanol/ton raw rice straw biomass	(Maibam & Goyal, 2025)
Rice husk	Pretreated with 1.5 M sodium hydroxide at a ratio of 1:10 (w/v) at 80℃ for 6 h and HPAC solution (1:1 (v/v) ratio of 30% (w/w) hydrogen peroxide and glacial acetic acid) at ratio of 1:10 (w/v), at 85℃ for 3 h. Pretreated rice husk hydrolyzed by xylose isomerase as well as D-psicose-3-epimerase with borate and fermented *S. cerevisiae* at 30℃ for 1 day	Ethanol with yields of approximately 141.4 g/kg rice husk biomass	(Song et al., 2024)
Rice husk and microalgal	Pretreated through acid-alkaline pretreatment by 1 N sodium hydroxide (NaOH) for 24 h and in 1.47% (v/v) sulfuric acid (H_2SO_4) solution Enzymatic saccharification of microalgal biomass hydrolysate and rice husk hydrolysate feedstock with 1:3 ratio by B. subtilis cell-free supernatant followed by fermentation by *K. marxianus* at pH 5.5 and 30℃ for 48 h	Ethanol yields of 140 mg ethanol/g biomass	(Mitra et al., 2025)
Rice husk and wheat husk	Pretreated through ionic liquid pretreatment by 1-ethyl-3-methylimidazolium acetate EMIM [OAc] at 100℃ for 3 h. The pretreated rice husk and wheat husk enzymatically hydrolyzed by purified MS2A cellulase and fermented by *S. cerevisiae*	Ethanol with yield of 23.43 g ethanol/L glucose from rice husk (RH) and 24.28 g ethanol/L glucose from wheat husk (WH)	(John & Selvarajan, 2024)
Sugarcane bagasse	Pretreated with 2.5% NaOH followed by steaming at 121℃. Saccharification of alkali-	Ethanol with yield of 0.49 g ethanol/g substrate	(Iram et al., 2018)

Feedstock	Operating Conditions	Performance	Ref.
	pretreated sugarcane bagasse by commercial cellulase enzyme (CM Case activity of 2900 IU/ml and filter paper activity of 1500 FPU/ml) and fermented at 30℃ by *S. cerevisiae*		
Sugarcane bagasse	Pretreated with 2.5% NaOH followed by steaming at 121℃. Saccharification of alkali-pretreated sugarcane bagasse by commercial cellulase enzyme (CM Case activity of 2900 IU/ml and filter paper activity of 1500 FPU/ml) and fermented at 30℃ by *P. stipites*	Ethanol with yield of 88.5 g/L ethanol	(Iram et al., 2018)
Sugarcane bagasse	Pretreated through 2 stages of salt-alkali pretreatment with 1.73 M $ZnCl_2$ and 1.36 M NaOH autoclaved for 30 min at 121℃ for both stages. SSF (Simultaneous Saccharification and Fermentation) by *S. cerevisiae* at 40℃ with 100 U/g enzyme loading	Ethanol with concentration of 4.88 g/L was produced	(Jugwanth et al., 2020)
Sugarcane bagasse	Deashing step was employed by utilizing citric acid trisodium dihydrate salt followed by autoclaving at 120℃ for 4 h and hydrolyzed using cerium-doped iron oxide ($CeFe_3O_4$) nanoparticles (NPs). Hydrolyzed feedstock fermented by *S. cerevisiae* (5%, v/v) at pH 5 and 30℃ for 24 h with malt extract (3.0 g/L), yeast extract (3.0 g/L), and peptone (5.0 g/L) included as nutrients	The ethanol concentration of 16.4 ± 1.68 g/L was achieved and the productivity was 1.36 ± 0.14 g/L/h	(A. Singh & Kim, 2025)
Sugarcane leaf	Pretreated with pretreated with 2%(w/v) of NaOH with solid to liquid ratio of 1:3 for 3 days. Hydrolyzed by commercial cellulase enzyme and β-glucosidase at ambient temperature (33–35 ℃) for 72h and fermented by 2% (w/v) of *S. cerevisiae* TISTR5020 at 35℃ for 96 h	Ethanol yield coefficient, ethanol productivity,fermentation efficiency and energy from ethanol were 0.141 ± 0.323 g/g, 0.109 ± 0.019 g/L·h, 27.687% and 157.020 ± 27.197 kJ/L, respectively were achieved	(Manmai et al., 2020)
Sugarcane trash (dry leaves, green leaves, and tops) and jatropha seed	Pretreated through alkaline pretreatment by sodium hydroxide (15 ml/g of liquid to solid ratio), 5% H_2O_2 (liquid to agrowaste, 20 ml/g), and Sodium hypochlorite (2–1 w/w). Then, subjected to saccharification by commercial cellulases (30 IU/g substrate) and fermented by *S. cerevisiae* F-307 at 34℃ for 72 h	The bioethanol yields were 325.4, 310.8, 282.9, 302.4 and 264.0 mg ethanol/g treated agrowastes from green leaves of sugarcane, jatropha deolied seed cake, tops sugarcane, dry leaves of sugarcane, and jatropha shell, respectively.	(Elnagdy et al., 2024)
Corn cob	Hydrolyzed with 13.1 M (70 %) concentration H_2SO_2 at a ratio of 1:5 (w/v) through 2 stages (first stage 100℃ for 60 min and second stage at 100℃ for 50 min). Fermented by *S.cerevisiae* at 30℃ for 72h	Ethanol with yield of 66.23 ± 8.35 mL/kg was obtained.	(Sokan-Adeaga et al., 2024)
Corn cob, eucalyptus, and cypress	Pretreated using 0.1M NaOH (w/v) and 0.1M HCl (v/v) at 60℃ for 2 h and at 121℃ (15 psi) for 15 min, then dried at 70℃.	Result showed cellulases of *Xylaria sp*. KM03 produced the highest	(Kamande et al., 2024)

Feedstock	Operating Conditions	Performance	Ref.
	Simultaneous hydrolysis and fermentation by cellulases created by 3 different strains of *Xylaria sp*. and *Nemania sp*	amounts of ethanol at 28.59 ± 1.58 mg/ml, *Nemania sp*. KM01 produced yields of 28.72 ± 3.82 mg/ml, whereas *Xylaria sp*. KM02 produced 25.75 ± 0.21 mg/ml.	
Corn cob	Deashing step was employed by utilizing citric acid trisodium dihydrate salt followed by autoclaving at 120℃ for 4 h and hydrolyzed using cerium-doped iron oxide ($CeFe_3O_4$) nanoparticles (NPs). Fermented by *S. cerevisiae* (5%, v/v) at 5 pH and 30℃ for 24 h with malt extract (3.0 g/L), yeast extract (3.0 g/L), and peptone (5.0 g/L) included as nutrients	Ethanol with concentrations of 28.9 ± 0.15 g/L was produced with corresponding productivities were 2.40 ± 0.15 g/L/h within 12 h period	(A. Singh & Kim, 2025)
Corn stalk	Pretreated by blended 1 g of corn stalk with 10.0 g [Ch][Gly] (3 wt%) in a 15 mL capped glass pressure tube, and the pretreatment conditions were 140℃ for 2 h. Hydrolyzed with an enzyme cocktail at 50℃ for 120 h with constant agitation and fermented by *S. cerevisiae* with 33 wt% yeast loading at 37℃ for 48 h	Ethanol with yield of 0.296 g/g feedstock and concentration of 8.5 g/L as achieved.	(Q. Zhu et al., 2023)
Corn stalk and leaves	Grounded feedstocks were added into alkaline solution of sodium hydroxide with different concentrations and 100 mL of nanoparticle solution at a solid loading, and then the mixture was autoclaved at different times (0, 15, 30 min). Then, hydrolyzed by commercial cellulase enzyme with a 2 % (v/v) enzyme loading for 24 h and then fermented by *S.cerevisiae* for 24 h	An increase in ethanol yield of 15.8 g/L in a fermentation period of 24 h was achieved with the pre-treatment at 2% NaOH with NiO NPs under the 15-minute autoclave condition	(Saetang & Tipnee, 2022)
Corn stalk	Pretreatment done by mixing Corn stalk, [BHEM] mesy, and ethylene glycol and were stirred at 140 ℃ for 2 h. Then through feed-batch method, rich-cellulose material, NaAc-HAc buffer solution and cellulast 1.5 L (160 EGU/g cellulose) were put into shaker at 200 rpm for 96 h under 50 ℃ at 4,8 pH for 96 h as an hydrolyzation process. Hydrolysate and yeast were mixed and stirred in shaker at 200 rpm for 48 h under 37℃, minimum yeast loading is 30 wt%	The ethanol concentration of 93 g/L with the yield of 73 % was achieved	(Q. Zhu et al., 2023)
Wheat husk	Dry biomass pretreated by of EMIM(OAC) for 3 h, heated to 100℃ and manually mixed every 10 min. The biomass was pre-treated, filtered, cleaned, and oven dried for an hour at 70℃. Hydrolyzed by 19U/ml of immobilized cellulase *A. niger* cellulase and *Streptomyces sp. MS2A cellulase* and further fermented by *S. cerevisiae.*	Production 26.18 ± 0.4 g/L and 20.06 ± 0.35 g/L of ethanol from *Streptomyces sp.* MS2A and *A. niger* hydrolysed product in wheat husk on the end of day 3.	(John J et al., 2026)
Wheat straw	Hydrolyzed in 2 stages. First, it was hydrolyzed with 10% (v/v) sulfuric acid at 100℃ for 20 min, then with 22% (v/v)	The process yielded optimal average bioethanol productivity	(P. Singh et al., 2025)

Feedstock	Operating Conditions	Performance	Ref.
	sulfuric acid concentration for 30 min at 100℃. Hydrolysate was fermented through constant volume fed-batch fermentation technique by *Z. mobilis* MTCC 91	(g/Lh), yield (g/g), and titer (g/L) of 7.147±0.533, 0.508±0.002, and 58.835±0.766; 5.722±0.529, 0.501±0.006, and 33.748±0.322	
Wheat straw	A sulfuric acid (1:10 biomass to acid ratio) of 10% (v/v) was used to hydrolyzed the wheat straw at 100℃ for 20 min. Hydrolysate was fermented in 0.5 L bioreactor by *Scheffersomyces stipitis* NCIM 3498	bioethanol with a yield and productivity of 0.4706±0.0022 g/g and 1.5325±0.0020 g/Lh; 0.4290±0.0030 g/g and 0.7399±0.0043 g/Lh, respectively was produced	(P. Singh et al., 2024)
Wheat straw	Pretreated with different solution such as and was heated for t = 60 minutes at a temperature T = 130ºC and a pressure of p = 3 bar. Hydrolyzed for 24 h under constant stirring and at 50ºC with the enzyme complex Accellerase 1,500 after being rinsed with water. Fermented by *S. cerevisiae* for 7 days	Wheat straw pretreated with KOH solution give the highest ethanol yield of 104.3 g/kg, while the lowest ethanol yield is 67.7 g/kg	(Tutt et al., 2012)
Barley straw	Pretreated in a steam explosion prototype maximum operating pressure of 4.12 MPa at 210℃ for 5 min. Through SSF, hydrolyzed with endo-xylanases cellulase, and β-glucosidase enzyme, and fermented by *K. marxianus* CECT 10875 at 42℃ for 72 h with 15% water insoluble solid.	Ethanol with highest yield of 29.4±0.3 g/L was obtained	(García-Aparicio et al., 2011)
Barley straw	Pretreated by the steam explosion in a 10 L reactor at 180℃ and 10 bar for 30 min. Hydrolyze by enzyme cocktail at pH 4.8 and 50℃ (0.05 M sodium citrate buffer) in an incubator shaker at 150 rpm. Pre-saccharification and SSF done through a pre-hydrolysis stage (8 h at 50℃) with a solid concentration of 20% (w/v) and an enzyme load (Cellic CTec2) of 30 FPU/g glucan, and fermentation stage at 35℃ using *S. cerevisiae* Ethanol Red for 48 h	12.6 g ethanol and 16.6 g lignin were obtained from 100 g of barley straw.	(Álvarez et al., 2021)
Oat straw	Pretreated by through 2 methods. First by Autohydrolysis where water was mixed with oat straw at liquid to solid ratio of 15g/g in a stirred stainless reactor at 205–220℃. Second with $Ca(OH)_2$ at 121℃ for 60 min, then extended to 90–134℃ for 30–240 min. SSF that was carried out by *S. cerevisiae* and enzymes CTec2 at 35℃, pH=5 and 150 rpm in an orbital incubator with water at liquid to solid ratio of 7and 20g/g using an enzyme loading of 10 and 20FPU/g.	Through both hydrolyzation process ethanol with concentration 50g/L was achieved	(Romaní et al., 2016)
Forage oat	Pretreated by γ-ray exposure from a 60 Co source at a dosage of 32 kGy for 2 h, mixed with organic acid mixture (lactic: acetic: propionic: butyric acid = 7.7: 2.3: 0.0: 0.0) and epiphytic microbiota, vacuum-sealed	Ethanol yield of 47.1 % was achieved	(Zhao et al., 2025)

Feedstock	Operating Conditions	Performance	Ref.
	again and stored at room temperature (27.5℃) for 60 days. pre-hydrolyzed for 48 h using cellulase (3000 U/g) and beta-glucosidase (300U/g). Fermented by *S. cerevisiae* strain (CICC 1308) with PBS at 30℃ with shaking at 150rpm for 144h,		
Oil palm empty fruit bunch (OPEFB)	Pretreated through acid–alkali pretreatment with a dilute acid solution (at substrate loading 12.50 %w/v, sulfuric acid concentration 0.2 M, at 121 ℃ for 53 min) followed by an alkali solution (NaOH 5 %w/v, at 121 ℃ for 20 min). Then, fermented through SSF at 30–45 ℃, 5–15% w/v substrate loading, pH 4–6, and 1–5%v/v yeast consentration.	Maximum yield of 0.281 g/g bioethanol yield was found at 12.24%w/v, pH 4.5, yeast concentration 2.04%v/v, and 36.94 ℃	(Sukhang, S., Choojit, S., Reungpeerakul, 2020)
Oil palm empty fruit bunch (OPEFB)	Pretreated by soaking it in 5 M NH4OH solution at 1:5 (weight to liquid) ratio for 24 h. Fermented through SSF on several concentrations with Cellulase enzyme Cellic Htec for the saccharification (50℃, 48 h, and 5 pH) and *Zymomonas mobilis* for the fermentation (for 24 h at 30℃)	The highest ethanol concentration is 0.25 g/L, obtained from fermenting 9% OPEFB loading in 3 h.	(Mardawati et al., 2019)
Oil palm empty fruit bunch (OPEFB)	Pretreated through acid-alkali pretreatment by with 2% (*v/v*) sulphuric acid (H_2SO_4) and 10% (*w/v*) sodium hydroxide (NaOH) solution. Next, SSF done by adding enzyme combination of 50 U/g of cellulase and 10 U/g of β-glucosidase and combination of microorganism (*S. cerevisiae* and *T. harzianum)* in a 50 mM citrate buffer (pH 4.8) at 50℃, 150 rpm for 72 h	The process produced 9.65 g/L of bioethanol, 0.46 g/g ethanol yield, and 89.56% conversion efficiency	(Derman et al., 2022)
Oil palm frond juice	Pretreated through 2 stages of Sseam explosion pretreatment in a 98 L capacity reactor with a maximum steam output of 12 bar (160℃, 4 bar, 30 min and 230℃, 10 bar, 4 min). SSF done by adding 3 wt% of CellicR CTec2 (Cellulase + β-glucosidase), incubated at 55 ℃ and shaken at 150 rpm for 72 hrs. Then, 10% (v/v) *S. cerevisiae* and 1% (v/v) yeast extract (10.0 g/l) were added and stirred at 150 rpm for 48 hrs.	The process roduce maxed bioethanol 33.89 g/l and 0.31 g ethanol/g glucose ethanol yield	(Srimachai et al., 2023)
Oil palm frond juice and fiber	Pretreated in microwave for 15 minutes water. Through, the juice hydrolyzed by CellicRCTec2 (Cellulase + β-glucosidase) 3% w/w (g-enzyme/g-cellulose) at 55℃ with shaking at 150 rpm for 96 h and fermented at 37 ℃ with shaking at 150 rpm for 24 h by adding 10% (v/v) of *S. cerevisiae* and 1% (v/v) yeast extract	The highest bioethanol yields at were in the range of 0.41-0.42 g-bioethanol/g-glucose.	(Srimachai & Phuket, 2024)
Oil palm trunk juice	Pretreated through 2 stages of Steam explosion pretreatment in a 98 L capacity reactor with a maximum steam output of 12 bar (160℃, 4 bar, 30 min and 230℃, 10 bar, 4 min). SSF done by adding 3 wt% of CellicR CTec2 (Cellulase + β-glucosidase), incubated at 55℃ and shaken at 150 rpm for 72 hrs.	The process produced maxed bioethanol 44.65 g/l and 0.40 g ethanol/g glucose ethanol yield	(Srimachai et al., 2023)

Feedstock	Operating Conditions	Performance	Ref.
	Then, 10% (v/v) *S. cerevisiae* and 1% (v/v) yeast extract (10.0 g/l) were added and stirred at 150 rpm for 48 hrs.		
Oil palm trunk sap	Sterilized by using steam at 120℃ before fermentation. The fermentation process was conducted using a batch strategy with pH of 6.50, a CSL (Corn steep liquor) concentration of 6.80 g/L, and a peptone concentration of 13.28 g/L as the optimal condition.	Bioethanol with yield of 0.4800 ± 0.0275 g/g and concentrations of 46.13 g/L was achieved	(Norhazimah et al., 2025)
Oil palm trunk sap and Oil palm trunk fiber (OPT)	The OPT fiber pretreated by 1% H_2SO_4 aqueous solution with 10% (*w/v*) the solid-to-liquid ratio at 121 ℃ for 30 min in autoclave. Five cycles of repeated-batch SSF done by adding concentrated sap and 10% *w/v* of OPT, 15 FPU/g OPT fiber of cellulase enzyme, 3 g/L of yeast extract, and at 30℃, 6 pH, and 150 rpm for 72 h.	Maximum ethanol production of 49.63 ± 1.05 g/L was achieved	(Billateh & Cheirsilp, 2024)
Oil palm mesocarp fiber and extract of Palm Empty Cluster Ash	Delignification was done by mixing fiber and palm empty fruit bunches ash extract with ration of 1:10 at 100℃ for 60 min followed by bleaching with mixing Fiber powder and H2O2 3% with ratio of 1: 5 90℃ for 60 min. The fiber powder then hydrolyzed with H_2SO_4 2 M with 1:20 ratio at 100℃ for 3 h. Then, fermented with *S. cerevisiae* on different concentrations.	The highest level of bioethanol concentration was obtained at 4 gr/L yeast variation of 7% (v/v) or 55.25 g/L at 96 h	(Ahmad et al., 2019)
Sorghum stalk	Pretreated in batch rector with varying concentrations of H_2SO_4 catalyst (0.45–1.35%), temperatures (90–130 ℃), and reaction times (60–120 min) for comparation. Then, SSF done in 2 L reactor where remaining solid residue was pre-digested at 50 ℃ with 25 FPU/g Cellic Ctec2, followed by 6 h of hydrolysis at 300 rpm and fermentation carried out by *S. cerevisiae* (No. TISTR 5339) at 35 ℃ and 150 rpm, and for 120 h.	The maximum ethanol concentration of 23.1 g/L occurred at 72 h under 25 FPU/g substrate at pH 4.8 and decreased 72 h after fermentation	(Kreetachat et al., 2025)
Sorghum stalk	Pretreated with pretreated with 2%(w/v) of NaOH with solid to liquid ratio of 1:3 for 3 days. Hydrolyzed by commercial cellulase enzyme and β-glucosidase at ambient temperature (33–35 ℃) for 72h and fermented by 2%(w/v) of *Saccharomyces cerevisiae* TISTR5020 at 35℃ for 96 h	Ethanol yield coefficient, ethanol productivity, fermentation efficiency and energy from ethanol of SS were 0.155 ± 2.177 g/g, 0.174 ± 0.038 g/L·h, 30.333% and 251.232 ± 54.393 kJ/L, respectively were achieved	(Manmai et al., 2020)
Sorghum stalk	Pretreated by Microwave-assisted deep eutectic solvent (DES) pretreatment using choline chloride-formic acid (ChCl: FA) in optimal conditions (ChCl: FA ratio of 1:3, 20 min, 250 W). Hydrolyzed by 10 FPU/g cellulase and fermented by *Saccharomyces cerevisiae* for 27 h	Maximum ethanol yield of 0.57 g/g was achieved	(Venkatachalam, 2025)
Sorghum straw	Pretreated and hydrolyzed by sodium hydroxide (NaOH) from 2% (w/v), and heating temperatures from 40 ℃ for 3 days.	Ethanol yield coefficient, ethanol productivity, fermentation efficiency	(Manmai et al., 2020)

Feedstock	Operating Conditions	Performance	Ref.
	Hydrolyzed by 2%(v/v) of cellulose enzyme at (33–35 ℃) and 5 pH for 72 h, and fermented by 2%(w/v) of yeast (*S. cerevisiae* TISTR5020) 35℃ for 96 h	and energy from ethanol were 0.155 ± 2.177 g/g, 0.174 ± 0.038 g/L·h, 30.333% and 251.232 ± 54.393 kJ/L, respectively was achieved	
Sorghum straw	Pretreated by sulfuric acid solution of 1% at 125 ℃ for 10 minutes in autoclaved and by 1.25 % sodium hydroxide with in an autoclave for 10 minutes at 90 ℃. Then, hydrolyzed with diluted H2SO4 of concentration 2%, 3% and 4% (v/v), temperature (110 ℃, 125 ℃ and 140 ℃), and time (20, 40 and 60) minutes and fermented by *S. cerevisiae* at 30 ℃ for 72 h with varying inoculum sizes (5%, 10%, and 15%) and pH (4.5, 5, and 5.5).	Maximum ethanol yield of 0.617 mL/g (48.742%) produced at a sugar content of 32.9875 g/L, pH of 5, and inoculum size of 15%	(Dube & Gebriheta, 2022)
Kenaf stems	Pretreated with alkali pretreatment by NaOH 2.5% and biological pretreatment by laccase enzyme (50U/g substrate) and 1-hydroxybenzotriazole (HBT) (5 % or 10 %) in 4.7 pH at 40℃ for 24 h at 100 rpm in an incubator shaker. Hydrolyzed by commercial cellulase (50 FPU) with Tween 20 as an additive in an incubator shaker at 50 ˚C for 72 h at 120 rpm. Fermented by 5% *S. cerevisiae* at 30 ℃ in an incubator shaker at 150 rpm for 48 h.	38.2 g/l of ethanol was obtained through the process	(L. Sharma et al., 2025)
Cotton stalk	Pretreated with 12.5 % of total solid load and 7.5 % of orthophosphoric acid at 121˚C. SSF done with the dosages of enzymes and yeast were 40 FPU g-1 of cellulase, 25 U ml-1 of xylanase and 10 % (v/v) respectively at 30˚C with 100 rpm for 96 h	Ethanol concentration of 32.65 g/L was achieved	(Sriramajayam et al., 2022)
Cotton stalk	Simultaneous pretreatment and saccharification Laccase done using cellulase enzyme cocktail with cellulase-to-Laccase ratio of 4:1 (v/v) at pH 5 and 50 ℃ for 24 h. Then, co-fermented with *Saccharomyces cerevisiae* for hexose sugars and *Pichia kudriavzevii* for pentose sugars at 33℃ for 72 h	The process produced maximum 47.75 ± 0.84 g/L ethanol with a yield of 0.45 g/g and productivity of 1.32 g/L/h. Fermentation produced maximum ethanol yield at 36 h	(S. Dey et al., 2026)
Cotton stalk	Pretreated by mixing it with sodium hydroxide (NaOH) at a concentration of 3.0% (w/v) at a solid loading of 2% (w/v) in microwave at 100℃ for 10 min. Hydrolyzed with 25 FPU/g- solid substrate Cellulase enzyme at pH of 5.0 with 2.0% (w/v) pretreated cotton stalk in incubator at 50℃ for 96 h with 120 rpm. Fermented through single and repeated batch simultaneous saccharification co-fermentation by immobilized *S. cerevisiae* and *P. tannophilus* at 30℃, 5 pH with 150 rpm for each cycle (96 h)	Ethanol production was the highest in the first batch cycle (9.21 g/L) with bioethanol concentration was gradually decreased and reached to a minimum level of 2 g/L at the 5^{th} cycle	(Malik et al., 2021)

Feedstock	Operating Conditions	Performance	Ref.
Sesame plant residue	Pretreated through Microbial pretreatments by *P. chrysosporium* for a period of 60 h at 30 ℃ at 150 rpm with shaker and by diluted sulfuric acid (1% H2SO4) for a period of 60 min at 100 ℃. Fermented in a sterilized bioreactor assisted with nitrogen aeration and agitation system, under 30 ℃ and 120 rpm agitation speed for a period of 60 h by 6 g·L−1 of *S. cerevisiae*	Maximum bioethanol yield of 1.90 g·L−1 was achieved	(Kumar et al., 2020)
Sunflower stalk	Pretreated by steam explosion at 15 psi for 1.5 h followed by sudden depressurization and filtered. Hydrolyzed by *T. reesei* Rut-C30 NRRL 11460 for 8 days at 28 ℃. Fermented under optimum conditions of fermentation time (24 h), pH (5.0), temperature (30 ℃) and inoculum size (3% v/v)	Maximum ethanol yield of 0.444 g/g was achieved.	(S.K. Sharma, K.L. Kalra, 2002)
Sunflower stalk	Hydrolyzed by soaking it in 0.02 M H_2SO_4 for overnight, was steam exploded at 207 ℃ and 21 kg/cm2 for 3 minutes. Then hydrolyzed at 50 ℃, pH 4.8 citrate buffer) with pulp/buffer 6% (w/w) and Celluclast 1.5L/pulp 2.67% (w/w). Fermented with at 30 ℃ and 3.2 pH with 80 rpm shaking for 24 h	Maximum ethanol yield of 0.028 g/100 g was achieved	(Vaithanomsat et al., 2009)
Ramie stalk	Pretreated with 4% NaOH and 0.02% anthraquinone-2-sulfonic acid sodium salt (AQSS) as catalyzer at 170 degrees C for 1 h. Fermented through fed-batch to 20% of solid substrate concentration	The ethanol concentration could reach 63 g/L	(F. Guo , W. Sun , X. Li , J. Zhao, 2014)
Groundnut shells and peels	Hydrolysis using H2SO4 of various concentrations (4% for peels and 6% for shells w/v). The hydrolysis was performed at varying reaction time (60 min for peel and 90 min for shells), and temperature of 100℃ for peels and 120℃ for shells. Fermented by *S. cerevisiae* for 5 days in 5.5 pH.	7.89% and 3.94% of ethanol were recovered from 100 g of peels and shells.	(Iyabode Bright & Olayinka, 2020)
Groundnut shells	The shells were dried and milled to100 microns. Hydrolyzed with 5% sulphuric acid solution at 900℃ for 2 h. Fermented at 25℃ for 3 days with Baker's yeast at ($50x10^5$ cells/1000mL)	90.62% v/v purity of ethanol was obtained, and yield was found to be 27.5%	(Nwaze et al., 2023)
Soybean hulls	Pretreated at 60 ℃ with aqueous H_2SO_4 (5mM) and midazole at 120 and 180 ℃ with 1 and 3 h of reaction time. Hydrolyzed at 4.8 pH, 50 ℃ for 48 h in an orbital shaker at 150 rpm with 1/9 ratio of Cellic HTec2®/Cellic CTec2® (*v/v*) and xylanases with enzyme loading of 20 FPU and 1206.7 U per gram of solid biomass, respectively. Fermented with *S. cerevisiae* for 12 h.	A maximum bioethanol yield of 78.9%	(Nishida et al., 2023)
Soybean hull (SH)	Pretreated through 2 steps acid pretreatment. Firstly, 3.00 mL of 72% *v/v* H_2SO_4 was added to 300.0 mg of ground SH and incubated at 30 ℃ for 1 h. Hydrolyzed by cellulase commercial enzyme at 50 ℃, 5.0 pH for 72 h.	An average ethanol yield of 0.45 $g_{ethanol}/g_{glucose}$ was achieved	(Gil Rolón et al., 2023)

Feedstock	Operating Conditions	Performance	Ref.
	Fermented by *S. cerevisiae* var. Ethanol red at 30 ℃ and stirring at 150 rpm for 12 h		

The analysis of the data given in Table 1 showed that cereal grasses and sugar-yielding crop residues able to give higher ethanol consentration and yield. This result comes from cell wall of the grass families which generally feature a less recalcitrant lignocellulosic matrix than rigid hardwood residues, making them highly susceptible to enzymatic breakdown (Sarkar et al., 2012). This result can be seen from a cereal grass-type feedstock such as corn stalks which give the highest reported bioethanol concentration in the dataset, reaching 93 g/L with a 73% yield via a [BHEM] mesy-ethylene glycol solvent pretreatment coupled with a fed-batch operational mode. Another type of feedstock that supports this statement can also be seen from sugarcane bagasse with bioethanol concentration reaching up to 88.5 g/L when fermented by *P. stipites* and sorghum stalk or straw which deliver maximum yields ranging from 0.57 g/g using the MAP-Deep Eutectic Solvents (DES) method to 0.617 mL/g under optimized conditions. Conversely, hard-shelled feedstock such as grounded nut shells or sesame residue produces significantly lower bioethanol concentration, usually only around 1.90 g/L or lower. This poor performance stems from a highly dense structural lignin shield enclosing the fiber matrix, which impedes enzymatic hydrolysis by standard cellulase enzymes due to its hydrophobic characteristic (Zeng et al., 2014). While, high-hemicellulose feedstocks such wheat straw usually demand strategic co-fermentation or genetically engineered strains to prevent pentose accumulation with bioconversion efficiency reaching only as high as 70%. Therefore, it can be concluded that feedstock with high cellulose/glucan content combined with a low or easily degradable lignin matrix, which is typical of the cereal grass and sugar-yielding crop family, are the best biomass composition for bioethanol production.

The high bioethanol concentration and yields result not only stem from the type of the feedstocks, but also from how these feedstocks are pretreated. The data shown that pretreatment by DES and Ionic Liquids (IL) resulting in higher bioethanol concentration and yield than other type of pretreatment method such as Alkali (NaOH) and Dilute Acid (H_2SO_4). For example, as choline chloride-based DES applied to sorghum stalks successfully unlocked a top-tier yield of 0.57 g/g, while the application of [BHEM] mesy on corn stalks yielded a peak concentration of 93 g/L. These pretreatments also combined with nanoparticle-assisted catalysts such as NiO NPs or magnetic $CeFe_3O_4$.

From the data analysis, it can also be concluded that *Saccharomyces cerevisiae* is the most common yeast strain used during fermentation in 75% of the compiled data due its low-cost, robust low-pH tolerance, superior ethanol toxicity resistance, and highly efficient metabolic pathways for converting hexose sugars like glucose. However, *S. cerevisiae* only able to ferment hexoses but not pentoses sugar which are what high-hemicellulose feedstocks, such as wheat straw, are consisted of. In order to ferment this kind of feedstock, another type of yeast such as *Pichia stipitis* and *Scheffersomyces stipitis* need to be added along with *Saccharomyces cerevisiae* to ferment both hexose and pentose sugars (Mohd Azhar et al., 2017).

Bioreactor Design and Fermentation Technologies

Conventional Bioreactor Configurations

Bioethanol production depends on conventional bioreactors that create stable and controlled conditions for microorganisms to convert fermentable sugars into ethanol efficiently. The most widely used reactor types are stirred tank reactors (STR), Bubble Column Reactors (BCR), and Airlift Reactors (ALR), each with their own flow characteristics and mass transfer efficiency (de Jesus et al., 2017; Suhaimi & Nasir, 2017). STR are generally favored because they provide intensive mixing, relatively uniform temperature distribution, and a high degree of operational flexibility, which supports their use in both batch and fed-batch fermentation schemes. However, their mechanical agitation can generate high shear forces which may damage shear-sensitive cells or microorganisms and typically require more energy for operation due to the mechanical components involved (de Jesus et al., 2017). The STR is widely used in fermentation and uses impellers for proper mixing and effective heat and mass transfer. Its temperature is maintained by a cooling jacket, while sterile air is supplied through a filtered line. The system also includes ports for adding nutrients, sampling, and collecting the product to maintain controlled microbial growth and efficient bioconversion like illustrate in Fig. 3.

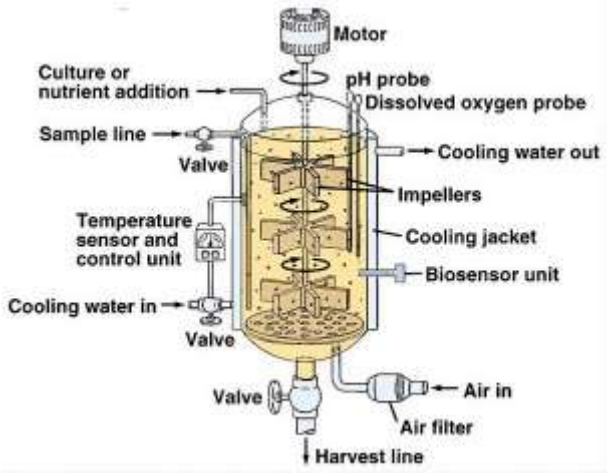

Figure 3. Design of Stirred Tank Reactor (STR) for Bioethanol Production (I. K. and A. D. Sharma, n.d.)

By contrast, pneumatically agitated reactors like BCR and ALR utilize gas induced circulation as the main mixing mechanism and are often selected for aerobic or co-culture fermentations, because they impose lower shear stress and require simpler mechanical components (Geng et al., 2021; Makki et al., 2023). Although generally have lower mixing efficiency compared to STR, which can lead to issues with scale-up, product quality, and process reproducibility. The gas-liquid interface in BCR and ALR can cause foaming, which may damage certain cells and complicate the process (Xu et al., 2022).

At laboratory and pilot scales, conventional bioreactors play a crucial role as experimental platforms for investigating microbial growth, ethanol yield, and fermentation kinetics. Critical process variables, including agitation speed, aeration rate, temperature, pH, and nutrient availability, must be carefully adjusted to maintain favorable conditions for yeast-based, bacterial, or enzyme assisted fermentation processes (Tyupa et al., 2021). Despite their long standing use and relatively straightforward scalability, these reactor systems still face several constraints, such as difficulties in achieving uniform substrate distribution, the accumulation of inhibitory compounds derived from biomass hydrolysates, and challenges in sustaining stable microbial performance, particularly when complex and heterogeneous lignocellulosic feedstocks are employed (Pino et al., 2018).

The selection of a suitable bioreactor depends not only on bioethanol productivity but also on the physicochemical properties of the fermentation broth, oxygen transfer requirements, substrate characteristics, and process scale. Stirred tank reactors remain the preferred option for most industrial bioethanol processes because of their operational flexibility and well-established scale-up methodology (Imamoglu & Sukan, 2013; Martín et al., 2018). In contrast, pneumatically agitated reactors are attractive for processes requiring reduced shear stress and lower energy consumption, although their mixing performance and gas–liquid mass transfer become increasingly challenging at larger scales (Geng et al., 2021).

Advanced Bioreactor Technologies

To address the shortcomings of conventional reactor systems, range of advanced bioreactor technologies has been developed with the objective of improving ethanol productivity, process efficiency, and operational stability. These systems include Packed Bed Reactor (PBR), Fluidized Bed Reactor (FBR), Membrane Bioreactor (MBR), and Continuous Stirred Tank Reactor (CSTR) that utilize immobilized microbial cells (Chandola & Agnihotri, 2021). PBR and FBR allow the retention of high cell concentrations and promote improved mass transfer, thereby enabling longer operating periods and higher ethanol production rates (Jang et al., 2020; Zapater et al., 2024). MBR further enhances process performance by enabling the selective removal of inhibitory compounds or ethanol during fermentation, which helps to reduce product inhibition. Such configurations are especially advantageous when processing lignocellulosic hydrolysates, typically exhibit variable sugar compositions and contain compounds that interfere with microbial activity (Jalilnejad et al., 2019).

In addition to these reactor configurations, immobilized-cell bioreactors have attracted considerable attention because they enable prolonged cell retention, repeated-batch operation, and continuous fermentation while minimizing biomass washout. Various carrier materials, including alginate beads, porous ceramics, activated carbon, and synthetic polymers, have been employed to improve microbial stability and ethanol productivity (Al-Qodah et al., 2018; Gao et al., 2021). Immobilized-cell systems are particularly

advantageous for lignocellulosic bioethanol production because they enhance microbial tolerance against inhibitory compounds present in biomass hydrolysates.

Beyond design innovations, these technologies often integrate intensification tactics like continuous modes, cell immobilization, and real time sensing of pH, temperature, and dissolved oxygen (DO). CSTR equipped with immobilized cells can provide greater fermentation stability, support continuous or repeated substrate feeding, and reduce the downtime commonly associated with batch operations (Y. F. Li et al., 2011). However, these advantages are accompanied by increased design complexity, more demanding control systems, and higher maintenance requirements (Horváthová et al., 2020). As a result, advanced bioreactor technologies are generally more suitable for laboratory scale and industrial scale applications, where improvements in efficiency, scalability, and long term operational reliability justify the additional technical demands. The key characteristics of conventional and advanced bioreactor configurations discussed above are summarized in Table 2. The comparison highlights their operating principles, major advantages, limitations, and typical applications in lignocellulosic bioethanol production, thereby providing a practical basis for selecting appropriate reactor systems according to process requirements.

Table 2. Comparison of Bioreactor Configurations for Lignocellulosic Bioethanol Production

Bioreactor	Mixing Mechanism	Advantages	Limitations	Suitability	Ref.
Stirred Tank Reactor (STR)	Mechanical agitation using impeller	- Excellent mixing and heat transfer - Homogeneous temperature and substrate distribution - Flexible operation (batch, fed-batch, continuous) - Well-established scale-up methodology - Compatible with enzymatic hydrolysis and microbial fermentation	- High energy consumption - High shear stress may affect shear-sensitive microorganisms - Impeller wear - Higher operational and maintenance costs - Particularly at high-solids loading	- Laboratory - Pilot-scale - Industrial fermentation of lignocellulosic hydrolysates - SHF and SSF processes	(Kadic & Heindel, 2010; Verardi et al., 2016)
Bubble Column Reactor (BCR)	Gas sparging and pneumatic mixing	- Simple reactor design - No moving mechanical parts - Low capital and maintenance costs - Low energy consumption - Reduced shear stress - Suitable for aerobic cultures	- Lower mixing efficiency than STR - Non-uniform substrate distribution at high solids loading - Foaming - Gas channeling - Limited mass transfer and scale-up performance	- Aerobic fermentation - Microbial cultivation - Co-culture systems - Low-viscosity lignocellulosic hydrolysates	(Ferreira et al., 2017; Makki et al., 2023)
Airlift Reactor (ALR)	Gas-induced liquid circulation through riser and downcomer	- Better liquid circulation than BCR - Low shear stress - Improved gas-liquid mass transfer - Lower power consumption	- Hydrodynamics strongly depend on reactor geometry - Lower mixing intensity than STR - Limited performance for highly viscous or high-solids	- Aerobic fermentation - Yeast cultivation - Immobilized-cell fermentation - Shear-sensitive microbial processes	(Valdivia-Rivera et al., 2019; J. Zhu et al., 2025)

Bioreactor	Mixing Mechanism	Advantages	Limitations	Suitability	Ref.
		- Suitable for sensitive microorganisms	lignocellulosic slurries		
Packed Bed Reactor (PBR)	Contonious flow through a packed bed containing immobilized cells	- High cell density - Prolonged cell retention - Excellent volumetric productivity - Enhanced microbial stability - Improved tolerance to inhibitory compounds - Reduced cell washout	- Channeling - Bed clogging due to suspended solids - Pressure drop - Difficult cleaning and carrier replacement	Continuous production using immobilized yeast or bacteria from clarified lignocellulosic hydrolysates	(C. W. Chen et al., 2023; Q. Liu et al., 2020; Sen et al., 2017)
Fluidized Bed Reactor (FBR)	Fluidization of immobilized carrier particles by upward liquids flow	- High mass transfer rates - Excellent mixing - Reduced clogging compared with PBR - High biomass loading - Improved substrate accessibility	- Complex hydrodynamics - Carrier particle attrition or loss - More difficult reactor control - Higher design complexity and capital cost	Continuous fermentation with immobilized microorganisms for lignocellulosic hydrolysates containing moderate suspended solids (SS)	(Grace, 2016; Guajardo et al., 2025)
Membrane Bioreactor (MBR)	Membrane-assisted separation integrated with information	- High cell retention - Reduced product inhibition - Improved fermentation efficiency - Facilitates cell recycling	- Membrane fouling - High membrane replacement cost - Increased maintenance - Pressure loss - Sophisticated process control required	Continuous fermentation coupled with product recovery or detoxification of lignocellulosic hydrolysates	(Ghifarhadi Prasiefa & Ali, 2026; Jefferson et al., 2018; Q. Wang & Cui, 2025)
Continuous Stirred Tank Reactor (CSTR)	Continuous mechanical agitation with simultaneous feed and product withdrawal	- Stable continuous production - High process automation - Constant operating conditions - Suitable for long-term industrial operation - Compatible with immobilized-cell systems - Higher space-time productivity than batch systems	- Cell washout without immobilization - More complex process control - Contamination risks during prolonged operation - Greater instrumentation requirements	Continuous lignocellulosic bioethanol production under SHF, SSF, and immobilized-cell fermentation processes	(Ghifarhadi Prasiefa & Ali, 2026; López-Caamal et al., 2024)

As shown in Table 2, no single bioreactor configuration is universally optimal for all bioethanol production processes. Reactor selection should therefore be based on feedstock characteristics, fermentation strategy, production scale, and economic considerations to achieve an appropriate balance between ethanol productivity, operational stability, and process efficiency. These considerations become increasingly important during scale-up, where hydrodynamic behavior, mass transfer, and process control significantly influence industrial performance.

Scale-Up Challenges and Operational Constraints

The transition from laboratory fermentation to industrial scale bioethanol production introduces a variety of technical and operational challenges that can significantly influence overall process performance and ethanol yield. Key issues include maintaining effective mixing and heat transfer, preventing localized depletion of substrates, and controlling shear stress levels that may negatively affect microbial viability, particularly in the case of shear sensitive yeast strains or non-conventional microorganisms (González-Gloria et al., 2024). At larger scales, variations in feedstock composition and hydrolysate quality can increase the formation of inhibitory compounds, making it necessary to continuously adjust operational parameters such as agitation intensity, aeration rate, and nutrient supplementation (Ghazali & Razak, 2021). Moreover, industrial scale operation requires advanced automation and process control to ensure uniform temperature, pH, and DO distribution throughout the reactor while managing continuous feeding and product withdrawal. Mechanical factors including impeller configuration, reactor geometry, and pumping capacity, further influence mass transfer efficiency and substrate accessibility (Celik et al., 2024; Rodrigues et al., 2024). Overcoming these challenges demands an integrated approach that combines bioprocess modeling, laboratory testing, and adaptive control strategies to ensure consistent and reliable performance of optimized fermentation systems and engineered microbial strains under commercial bioethanol production conditions.

Successful industrial implementation also depends on appropriate scale-up strategies that preserve hydrodynamic similarity and fermentation performance across different reactor sizes. Common scale-up criteria include maintaining constant power input per unit volume (P/V), impeller tip speed, mixing time, or oxygen transfer coefficient (k_La), depending on the characteristics of the fermentation system. More recently, computational fluid dynamics (CFD) and digital process modeling have been increasingly employed to predict fluid flow, substrate gradients, heat transfer, and shear distribution, thereby supporting reactor optimization before pilot- and industrial-scale implementation (M. Chen & Xia, 2024; Z. Li et al., 2023).

Process Integration in Bioethanol Production

Coupling Between Hydrolysis and Fermentation Processes

Integrating hydrolysis and fermentation stands out in bioethanol workflows, especially via simultaneous saccharification and fermentation (SSF) or SSCF. Under these configurations, lignocellulosic biomass that has undergone pretreatment using diluted acids such as H_2SO_4 and hydrochloric acid (HCl) or alkaline agents including sodium hydroxide (NaOH) and calcium hydroxide ($Ca(OH)_2$) are processed within a single reactor where both enzymatic and microbial reactions take place at the same time (Lorenci Woiciechowski et al., 2020). During operation, hydrolytic enzymes convert cellulose and hemicellulose into fermentable sugars, which are immediately assimilated by fermentative microorganisms, including yeasts or genetically modified bacteria, for ethanol production. This integrated configuration minimizes the accumulation of free sugars that may otherwise inhibit enzymatic activity or microbial metabolism, thereby contributing to improved ethanol yield and overall process performance (Palupi et al., 2023).

The effective operation of SSF or SSCF depends on precise instrumentation and careful process control. STR are commonly employed and equipped with systems for temperature and pH regulation, agitation control, DO measurement, and online monitoring of sugar or ethanol concentrations. Maintaining enzyme activity and microbial viability requires careful addition of buffers, nutrients, and hydrolysis related reagents. Through the coordinated management of these operational variables, SSF and SSCF facilitate continuous substrate conversion, simplify process flows, and establish a synergistic operational environment in which hydrolysis and fermentation are effectively integrated within a single processing unit (Unyay et al., 2024).

Operational Synchronization for Enhanced Process Efficiency

Operational synchronization in bioethanol production refers to the coordinated management of process parameters across different stages to enhance efficiency and reduce energy demand. In contrast to the physical integration discussed in Section 6.1, this approach emphasizes the temporal alignment of key variables such as substrate feeding rates, agitation intensity, aeration, temperature, and pH control. Dynamic

real-time optimization (DRTO) approaches, such as those using genetic algorithms (GA) and differential evolution (DE), have been shown to optimize reactor feed rates effectively, considering process disturbances (De Freitas et al., 2017). Agitation and aeration are essential for maintaining homogeneous conditions and adequate oxygen transfer in aerobic fermentations (Lis et al., 2020). Another studies shown that maintaining an optimum temperature around 31℃ for *Saccharomyces cerevisiae* minimizes Gibbs free energy and maximizes fermentation efficiency (Aslan et al., 2025). The use of automated monitoring systems and real time control strategies can help maintain optimal pH levels, which maintaining the microbial and enzyme activity (Rivera et al., 2009). Proper synchronization ensures a steady supply of fermentable sugars, stable microbial activity, and timely alignment between ethanol formation and downstream processing, all of which are critical for maintaining favorable reaction kinetics throughout the production cycle.

Modern synchronization strategies increasingly incorporate automated feedback control systems, staged or controlled substrate feeding, and adaptive monitoring tools that respond to fluctuations in feedstock characteristics, enzyme performance, or microbial behavior. Artificial Intelligent (AI) and Machine Learning (ML) models enhance process optimization by predicting optimal conditions and improving enzyme selection. These technologies facilitate real-time monitoring and adaptive control, significantly improving process efficiency and scalability (Ananda et al., 2025; Machado et al., 2025; Srivastava, 2025; Thangavelu, 2026). By promoting a balanced and responsive interaction between hydrolysis, fermentation, and downstream operations, operational synchronization plays a central role in maximizing bioethanol output while maintaining product consistency and industrial scale process stability.

Industrial Implementation Challenges and Future Perspectives

Techno-Economic Analysis of Bioethanol Based Lignocellulosic Agricultural Waste

The main barriers of industrial lignocellulosic bioethanol implementation come from the high cost of lignocellulosic ethanol production. Enzymes is one of the most expensive inputs in lignocellulosic ethanol production. One of the reports estimate the cost of cellulase can reach as high as USD 0.45/gallon of ethanol, which is still considered to expensive for commercial use (Hermiati et al., 2010). Other factor that could influence the production economics of lignocellulosic bioethanol is the pretreatment process. One of the recent studies reported that pretreatment through deep eutectic solvent pretreatment reported a minimum ethanol selling price of US$ 2,128.1/ton for the ChCl-lactic acid ($C_3H_6O_3$) system, whereas the ChCl-urea ($(NH_2)_2CO$) system reached US$ 3,049.9/ton. This result showed how the importance of choosing the most effective and low-cost pretreatment methods to achieve lower Minimum Ethanol Eelling Price (MESP) (Peng et al., 2021).

In order to reduce the cost of both processes, an optimization of both pretreatment and enzyme loading need to be done. One study on bioethanol production from brown sugar palm solid waste showed that using 5% H_2SO_4 plus organo-solvent pretreatment, followed by cellulase from *Trichoderma reesei* and xylanase during hydrolysis can produce bioethanol with yield reaching 0.26%. This result illustrate that industrially relevant routes often require both chemical pretreatment and multiple enzymes with certain amount of loading to achieve efficient sugar release, which resulting in higher yield (Nury et al., 2024).

Higher yield with lower production cost can also be achieve by lowering the process energy consumption through cogeneration. One study showed how production of bioethanol from fruit bunch feedstock yielded 313.83 L ethanol/ton of dry biomass, with a production cost of 0.49 US$/L when cogeneration was included and 0.58 US$/L without cogeneration. This result showed that commercial feasibility also depends on how much process energy can be internally recovered and how effectively the plant is integrated with utility generation (Quintero et al., 2013).

Life Cycle Assessment (LCA)

The goal of LCA is to compare and evaluate lignocellulosic bioethanol's environmental performance relative to fossil gasoline or to compare different biomass pathways to each other, using 1 MJ of ethanol delivered or 1 kg of ethanol produced to allow energy-equivalent comparison (Amin et al., 2026; Morales-Vera et al., 2022). The system boundary ranges from cradle-to-gate, ending at the biorefinery gate, to cradle-to-grave, which extends through fuel distribution and combustion in a dedicated vehicle (Morales-Vera et al., 2022). The first step of LCA is to analyze the input and output of the process. For example, growing poplar trees for biofuel uses about 3,590,012 BTU of diesel and 5,239 grams of herbicide/hectare each year. Biomass is then transported (often around 100 km) to the biorefinery, where chemicals like H_2SO_4, ammonia (NH_3), and corn steep are used during pretreatment and fermentation process (Morales-Vera et al., 2022). For lignocellulosic feedstocks, cellulose, hemicellulose, and lignin contents vary by biomass type. According to a study, bagasse with 33–36% cellulose, 28–30% hemicellulose, and 17–24% lignin versus rice

straw with 32–47% cellulose, 19–27% hemicellulose, and 5–24% lignin. This different biomass composition directly influencing conversion efficiency and downstream inventory flows (Amin et al., 2026). Another study showed that in dimethyl imidazolidinone (DMI) solvent-based process, the amount of solvent needed during process can decreases from 3,456.7 kg/h down to just 84.0 kg/h after optimization, which also reduced the heat needed for recovery (Yang et al., 2026). Annually, large-scale bioethanol plant required 1.17 million L of fresh water usage for, compared to 2.87 times more without recirculation, resulting in a grey water footprint of 119,000 tons per year (Afedzi et al., 2025).

The known number above will be turn into environmental impact scores such as global warming potential (GWP), greenhouse gas (GHG) emissions and energy performance. Lignocellulosic bioethanol generally achieves 40–91% GHG reduction relative to gasoline, with GWP ranging from roughly 25.7 to 605 g CO_2-eq/MJ depending on feedstock and pretreatment method, such as 402 g CO_2-eq/MJ for DES-lactic acid-pretreated rice straw and 604 g CO_2-eq/MJ for DES-urea (Amin et al., 2026). Energy performance is commonly expressed as net energy ratio, which exceeds 1.5 for well-integrated waste/residue systems but falls below unity for less optimized lignocellulosic or algal pathways (Amin et al., 2026). In another case study, bioethanol from hybrid poplar showed a net GWP of -1.05×10^{-3} kg CO_2-eq/MJ; while driven by 2.6×10^{-2} kg CO_2-eq/MJ from feedstock production; 2.4×10^{-1} kg CO_2-eq/MJ from bioconversion; and 6.7×10^{-2} kg CO_2-eq/MJ from fuel use, offset by -3.1×10^{-1} kg CO_2-eq/MJ in carbon sequestration credits. By comparation, gasoline has higher carbon footprint's, reaching at 88 g CO_2/MJ (Morales-Vera et al., 2022). For DMI solvent-based process, GWP decrease from 128.7 kg CO_2-eq/kg ethanol to 12.0 kg CO_2-eq/kg after switching to leftover lignin or wood as heat source to decrease or substitute fuel. MESP fell correspondingly from \$11.0/kg to \$1,169.7/t from these optimization steps (Yang et al., 2026).

From the data of these various studies, it could be concluded that no single part of the process is responsible for all the environmental impact. Bioconversion contributed an average of 77% of environmental for poplar-based ethanol, ranging from 50% for smog to 97% for ozone depletion, while feedstock production was the second-largest contributor. To check this uncertainty, researchers run around 5,000 simulations on different scenarios in which it is found that there was about a 70% chance the real-world CO2 emissions would come in lower than their main estimate, which gives more confidence in the results, and 69.2% probability for the ethanol yield target of 275 L/dry tonne (Morales-Vera et al., 2022). Across these different studies, it can be interpretated that GHG reductions of up to 90% are achievable with optimized enzyme dosing and process integration. However, the different boundaries, units, and ways of crediting byproducts from these various studies become the primary barrier to drawing consistent conclusion about lignocellulosic bioethanol sustainability (Afedzi et al., 2025; Amin et al., 2026).

Integration with Biorefinery and Circular Economy

The concept of an integrated biorefinery is crucial for various conversion routes in order to optimized and maximized the bioconversion of lignocellulosic biomass into bioethanol with near-zero waste discharge (Rajesh Banu et al., 2021). The integration is through various pathways such as biochemical, thermochemical, physical, and catalytic pathways to minimize energy demands, reduce reaction times, and enhance overall process economics in a closed-loop system (Rajesh Banu et al., 2021; Velvizhi et al., 2022). Within this framework, utilizing biological agents such as fungi, bacteria, and ligninolytic enzymes will serve as an eco-friendly method for delignification, liberating cellulose and hemicellulose fractions (Liguori & Faraco, 2016). Economic analyses indicate that co-producing high-value side products alongside biofuels is far more profitable than producing fuels alone (Rajesh Banu et al., 2021). For instance, fractionating and valorizing lignin into functional polymers like polyurethane and polyester, rather than treating it as waste, significantly boosts profitability (Velvizhi et al., 2022). This economic efficiency is further demonstrated by one-pot fractionation processes that co-produce furfural, lignin, and ethanol, cutting furfural production costs by up to 37.5% below market price (Rajesh Banu et al., 2021).

Additionally, integrating biological routes diversifies the product portfolio by generating valuable commercial chemicals such as organic acids (lactic and succinic acids), biohydrogen, biomethane, PHA/PHB, and quercetin. Any remaining solid residues are not discarded but are recycled as agricultural soil conditioners or fuel briquettes. Ultimately, transitioning from a linear economy to a sustainable circular bioeconomy through the efficient utilization of agro-industrial wastes and perennial crops improves techno-economic viability, sharply mitigates greenhouse gas emissions, and reduces global dependence on fossil resources (Liguori & Faraco, 2016; Rajesh Banu et al., 2021; Velvizhi et al., 2022). However, to fully realize this circular bioeconomy model, the inevitable generation of inhibitory compounds during biomass pretreatment remains a critical bottleneck that requires thorough future evaluation. Severe pretreatment conditions often trigger the degradation of sugars and lignin into toxic by-products, including furans (such as furfural and HMF), phenolics, and weak organic acids (Guo et al., 2022). Because these compounds

severely impair enzymatic hydrolysis and downstream microbial fermentation, developing cost-effective biological detoxification methods and engineering robust, inhibitor-tolerant strains must be prioritized in upcoming research to ensure overall system viability

Conclusion

Lignocellulosic bioethanol production is strongly governed by the interactions among feedstock composition, pretreatment efficiency, hydrolysis performance, microbial metabolism, fermentation strategy, bioreactor configuration, and downstream processing. Feedstocks with higher cellulose content and lower lignin recalcitrance generally provide greater fermentable sugar availability, while advances in green pretreatment technologies, integrated saccharification-fermentation processes, engineered microorganisms, and optimized bioreactor designs have substantially improved ethanol productivity and overall process efficiency. Despite these technological advances, economic competitiveness is still constrained by high pretreatment and enzyme costs, inhibitor formation, energy consumption, water demand, and challenges associated with industrial-scale implementation.

Future research should prioritize the development of integrated biorefinery systems that combine sustainable pretreatment technologies, robust lignocellulose-fermenting microorganisms, artificial intelligence-assisted process optimization, continuous and intensified bioprocesses, and advanced reactor designs. Greater emphasis should also be placed on pilot-scale validation, solvent and water recycling, techno-economic assessment, life cycle assessment, and circular bioeconomy integration to bridge the gap between laboratory-scale achievements and commercial deployment. These multidisciplinary strategies are expected to improve production efficiency, reduce environmental impacts and operating costs, and accelerate the large-scale commercialization of lignocellulosic bioethanol as a sustainable transportation fuel.

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References

Afedzi, A. E. K., Afrakomah, G. S., Gyan, K., Khan, J., Seidu, R., Baidoo, T., Sultan, I. N., Tareen, A. K., & Parakulsuksatid, P. (2025). *Enhancing Economic and Environmental Sustainability in Lignocellulosic Bioethanol Production: Key Factors, Innovative Technologies, Policy Frameworks, and Social Considerations*. *17*(2), 499. https://www.scopus.com/pages/publications/85215823902?origin=scopusAI

Afedzi, A. E. K., & Parakulsuksatid, P. (2023). Recent advances in process modifications of simultaneous saccharification and fermentation (SSF) of lignocellulosic biomass for bioethanol production. *Biocatalysis and Agricultural Biotechnology*, *54*. https://doi.org/10.1016/j.bcab.2023.102961

Ahmad, A., Muria, S. R., & Tuljannah, M. (2019). Production of Second Generation Bioethanol from Palm Fruit Fiber Biomass using Saccharomyces cerevisiae. *Journal of Physics: Conference Series*, *1295*(1). https://doi.org/10.1088/1742-6596/1295/1/012030

Al-Qodah, Z., Al-Shannag, M., Al-Bosoul, M., Penchev, I., Al-Ahmadi, H., & Al-Qodah, K. (2018). On the performance of immobilized cell bioreactors utilizing a magnetic field. *Reviews in Chemical Engineering*, *34*(3), 385–408. https://doi.org/10.1515/revce-2016-0059

Álvarez, C., González, A., Ballesteros, I., & Negro, M. J. (2021). Production of xylooligosaccharides, bioethanol, and lignin from structural components of barley straw pretreated with a steam explosion. *Bioresource Technology*, *342*(September). https://doi.org/10.1016/j.biortech.2021.125953

Amin, M. M., Elbager, M. A., Ibrahim, R. H., Abdul Jameel, A. G., Razzak, S. A., & Siddiquee, M. N. (2026). Bioethanol revolution: Integrating feedstock innovation with energy-efficient processes under life cycle assessment. *Biomass Futures*, *2*, 100034. https://doi.org/10.1016/J.BMF.2026.100034

Ananda, A., Sujeeth, R. K., & Archana, S. (2025). Integrating Automation in Biomass Transformation: Opportunities, Challenges, and Future Directions. *Bioenergy Research*, *18*(1). https://doi.org/10.1007/s12155-025-10864-6

Aslan, C., Devianto, H., Wonoputri, V., & Harimawan, A. (2025). Study of cell elemental balance and cell thermodynamics in microorganism black box modeling: Prediction of bioethanol fermentation stoichiometry by Saccharomyces cerevisiae as a function of temperature. *AIP Conference Proceedings*, *3295*(1). https://doi.org/10.1063/5.0269556

Bacquerié, C., Guigou, M., Cebreiros, F., Larnaudie, V., Roman, M. E., Cagno, M., Rodríguez, F., Bonfiglio, F., Ferrari, M. D., & Lareo, C. (2025). Enhanced cellulose enzymatic hydrolysis of pilot-scale steam-exploded Eucalyptus grandis chips with previous acid impregnation for bioethanol production. *Energy Conversion and Management: X*, *27*. https://doi.org/10.1016/j.ecmx.2025.101197

Bae, J. H., Kim, M. J., Sung, B. H., Jin, Y. S., & Sohn, J. H. (2021). Directed evolution and secretory expression of xylose isomerase for improved utilisation of xylose in Saccharomyces cerevisiae. *Biotechnology for Biofuels*, *14*(1). https://doi.org/10.1186/s13068-021-02073-y

Bellido, C., Lucas, S., González-Benito, G., García-Cubero, M. T., & Coca, M. (2018). Synergistic positive effect of organic acids on the inhibitory effect of phenolic compounds on Acetone-Butanol-Ethanol (ABE) production. *Food and Bioproducts Processing*, *108*, 117–125. https://doi.org/10.1016/j.fbp.2018.02.004

Billateh, A., & Cheirsilp, B. (2024). Efficient Biovalorization of Oil Palm Trunk Waste as a Low-Cost Nutrient Source for Bioethanol Production. *Energies* , *17*(13). https://doi.org/10.3390/en17133217

Bioenergy Research Group. (n.d.). Retrieved February 17, 2026, from https://www2.hawaii.edu/~khanal/fungal/biofuels.html

Biotechnology in China III: Biofuels and Bioenergy - Google Buku. (n.d.). Retrieved February 17, 2026, from https://books.google.co.id/books?hl=id&lr=&id=p7e5BQAAQBAJ&oi=fnd&pg=PR3&dq=Biotechnology+in+China+III:+Biofuels+and+Bioenergy&ots=5vsqIsXDRO&sig=03qfJsXtGUUy5XgIEmzKgzRodaQ&redir_esc=y#v=onepage&q=Biotechnology in China III%3A Biofuels and Bioenergy&f=false

Camargos, C. V., Moraes, V. D., de Oliveira, L. M., Guidini, C. Z., Ribeiro, E. J., & Santos, L. D. (2021). High Gravity and Very High Gravity Fermentation of Sugarcane Molasses by Flocculating Saccharomyces cerevisiae: Experimental Investigation and Kinetic Modeling. *Applied Biochemistry and Biotechnology*, *193*(3), 807–821. https://doi.org/10.1007/s12010-020-03466-9

Caro, I., Marzo-Gago, C., Díaz, A. B., & Blandino, A. (2024). Modelling and optimization of simultaneous saccharification and fermentation of agro-food residues. *Journal of Environmental Chemical Engineering*, *12*(1). https://doi.org/10.1016/j.jece.2023.111862

Celik, A. F., Elibol, E. A., Turgut, O., Senol, H., & Sillanpää, M. (2024). Interpretation of possible biogas production capacity by investigating the effects of anaerobic digester tank geometry and angular velocity on flow characteristics. *Environmental Science and Pollution Research*, *31*(50), 60220–60234. https://doi.org/10.1007/s11356-024-35205-6

Chandola, D., & Agnihotri, V. (2021). Recent Perspectives of Immobilized Enzyme Reactors Used for Wastewater Treatment. In *Bioremediation of Environmental Pollutants: Emerging Trends and Strategies* (pp. 275–293). Springer International Publishing. https://doi.org/10.1007/978-3-030-86169-8_12

Chen, C. W., Mirzaei, S., Huang, C. C., & Li, S. Y. (2023). A scale-up study of the continuous ABE fermentation in a packed bed coupled with the extraction/gas-stripping in situ butanol recovery process. *Separation and Purification Technology*, *318*. https://doi.org/10.1016/j.seppur.2023.123952

Chen, M., & Xia, J. (2024). Optimization parameters for efficient scale-up of fermentation process. In *Scale-up and Chemical Process for Microbial Production of Plant-Derived Bioactive Compounds: From Laboratory to Industry* (pp. 29–42). Elsevier. https://doi.org/10.1016/B978-0-443-15584-0.00002-1

Cheng, C., Bao, T., & Yang, S. T. (2019). Engineering Clostridium for improved solvent production: recent progress and perspective. *Applied Microbiology and Biotechnology*, *103*(14), 5549–5566. https://doi.org/10.1007/s00253-019-09916-7

Chung, J. H., Lee, J., Kim, S., & Kim, K. H. (2025). Comparative Metabolomics of Clostridium acetobutylicum ATCC824 and its Engineered Strain, C. acetobutylicum DG1. *Journal of Microbiology and Biotechnology*, *35*. https://doi.org/10.4014/jmb.2407.07028

Coimbra, M. C., Duque, A., Saéz, F., Manzanares, P., Garcia-Cruz, C. H., & Ballesteros, M. (2016). Sugar production from wheat straw biomass by alkaline extrusion and enzymatic hydrolysis. *Renewable Energy*, *86*, 1060–1068. https://doi.org/10.1016/j.renene.2015.09.026

Cui, J., Maloney, M. I., Olson, D. G., & Lynd, L. R. (2020). Conversion of phosphoenolpyruvate to pyruvate in Thermoanaerobacterium saccharolyticum. *Metabolic Engineering Communications*, *10*. https://doi.org/10.1016/j.mec.2020.e00122

De Freitas, H. F. S., Olivo, J. E., & Andrade, C. M. G. (2017). Optimization of bioethanol in silico production process in a fed-batch bioreactor using non-linear model predictive control and evolutionary computation techniques. *Energies*, *10*(11). https://doi.org/10.3390/en10111763

de Jesus, S. S., Moreira Neto, J., & Maciel Filho, R. (2017). Hydrodynamics and mass transfer in bubble column, conventional airlift, stirred airlift and stirred tank bioreactors, using viscous fluid: A comparative study. *Biochemical Engineering Journal*, *118*, 70–81. https://doi.org/10.1016/j.bej.2016.11.019

Derman, E., Abdulla, R., Marbawi, H., Sabullah, M. K., Gansau, J. A., & Ravindra, P. (2022). Simultaneous Saccharification and Fermentation of Empty Fruit Bunches of Palm for Bioethanol Production Using a Microbial Consortium of S. cerevisiae and T. harzianum. *Fermentation*, *8*(7). https://doi.org/10.3390/fermentation8070295

Dessie, W., Xiao, J., Tang, J., An, B., Luo, X., Wang, M., Liao, Y., Wahab, R., Li, C., & Qin, Z. (2024). Maximizing fermentable feedstocks from Camellia oleifera seed oil extraction residues: Green pretreatment and enzymatic hydrolysis for effective valorization. *Arabian Journal of Chemistry*, *17*(6). https://doi.org/10.1016/j.arabjc.2024.105815

Dey, H., Paul, S., & Kaira, M. (2025). Metabolic/Genetic Engineering Approaches for Lignocellulosic Biomass and Enzymes. In *Lignocellulosic Biomass and Enzymes: Fundamentals, Emerging Technologies and Applications* (pp. 55–84). Springer Science+Business Media. https://doi.org/10.1007/978-981-96-3037-0_3

Dey, S., Banerjee, S., & Banerjee, R. (2026). Biotransformation of cotton stalk to bioethanol by an environmentally benign enzymatic route. *Biomass and Bioenergy*, *208*(August 2025), 108802. https://doi.org/10.1016/j.biombioe.2025.108802

Dube, A. M., & Gebriheta, F. G. (2022). *Bioethanol Production from Sequential Acidic-Alkaline Pretreated Sorghum Straw Hydrolysate and Investigation of the Effects of Fermentation Parameters using Response Surface Methodology*. (September), 0–23. https://doi.org/10.20944/preprints202209.0389.v1

Dutta, S., & Suresh Kumar, M. (2023). Potential use of thermophilic bacteria for second-generation bioethanol production using lignocellulosic feedstocks: a review. *Biofuels*, *14*(8), 851–864. https://doi.org/10.1080/17597269.2023.2184935

Elhalis, H. (2024). Expanding the Horizons of Saccharomyces cerevisiae: Nutrition, Oenology, and Bioethanol Production. *Sustainability (Switzerland)*, *16*(24). https://doi.org/10.3390/su162411151

Elnagdy, N. A., Ragab, T. I. M., Fadel, M. A., Abou-Zeid, M. A., & Esawy, M. A. (2024). Bioethanol Production from Characterized Pre-treated Sugarcane Trash and Jatropha Agrowastes. *Journal of Biotechnology*, *386*(December 2023), 28–41. https://doi.org/10.1016/j.jbiotec.2024.02.015

F. Guo , W. Sun , X. Li , J. Zhao, and Y. Q. (2014). Pretreatment of ramie and kenaf stalk for bioethanol production. *Sheng Wu Gong Cheng Xue Bao*, *30*(5), 774–783.

Ferreira, A., Rocha, F., Mota, A., & Teixeira, J. A. (2017). Nonmechanically Agitated Bioreactors. *Current Developments in Biotechnology and Bioengineering: Bioprocesses, Bioreactors and Controls*, 217–233. https://doi.org/10.1016/B978-0-444-63663-8.00008-2

Frohman, C. A., & Mira de Orduña, R. (2013). Cellular viability and kinetics of osmotic stress associated metabolites of Saccharomyces cerevisiae during traditional batch and fed-batch alcoholic fermentations at constant sugar concentrations. *Food Research International*, *53*(1), 551–555. https://doi.org/10.1016/j.foodres.2013.05.020

Gao, H., Lu, J., Jiang, Y., Fang, Y., Tang, Y., Yu, Z., Zhang, W., Xin, F., & Jiang, M. (2021). Material-mediated cell immobilization technology in the biological fermentation proces. *Biofuels, Bioproducts and Biorefining*, *15*(4), 1160–1173. https://doi.org/10.1002/bbb.2219

García-Aparicio, M. P., Oliva, J. M., Manzanares, P., Ballesteros, M., Ballesteros, I., González, A., & Negro, M. J. (2011). Second-generation ethanol production from steam exploded barley straw by Kluyveromyces marxianus CECT 10875. *Fuel*, *90*(4), 1624–1630. https://doi.org/10.1016/j.fuel.2010.10.052

Geng, S., Mao, Z. S., Huang, Q., & Yang, C. (2021). Process Intensification in Pneumatically Agitated Slurry Reactors. *Engineering*, *7*(3), 304–325. https://doi.org/10.1016/j.eng.2021.03.002

Ghazali, N. F., & Razak, N. D. A. (2021). Recovery of saccharides from lignocellulosic hydrolysates using nanofiltration membranes: A review. *Food and Bioproducts Processing*, *126*, 215–233. https://doi.org/10.1016/j.fbp.2021.01.006

Ghifarhadi Prasiefa, M. ', & Ali, M. N. (2026). Biogas Production from Livestock Manure via Anaerobic Digestion and Co-Digestion: A Comprehensive Review of Processes, Microbial Roles, Technological Perspectives, and Opportunities. *Journal of Biobased Chemicals*, *6*(1), 22–46. https://doi.org/10.19184/JOBC.V6I1.60002

Gil Rolón, M., Leonardi, R. J., Bolzico, B. C., Seluy, L. G., Benzzo, M. T., & Comelli, R. N. (2023). Multi-Response Optimization of Thermochemical Pretreatment of Soybean Hulls for 2G-Bioethanol Production. *Fermentation*, *9*(5). https://doi.org/10.3390/fermentation9050454

González-Gloria, K. D., Tomás-Pejó, E., Amaya-Delgado, L., Rodríguez-Jasso, R. M., Loredo-Treviño, A., Singh, A., Hans, M., Martín, C., Kumar, S., & Ruiz, H. A. (2024). Biochemical and Biorefinery Platform for Second-Generation Bioethanol: Fermentative Strategies and Microorganisms. *Fermentation*, *10*(7). https://doi.org/10.3390/fermentation10070361

Gou, Z., Li, Y., Xie, C., Tang, Y., & Kida, K. (2015). Evaluation of the inhibitor-tolerance of industrial Saccharomyces cerevisiae strain KF-7. *Chinese Journal of Applied and Environmental Biology*, *21*(2), 248–255. https://doi.org/10.3724/SP.J.1145.2014.08009

Grace, J. R. (2016). Fluidized-bed catalytic reactors. In *Multiphase Catalytic Reactors: Theory, Design, Manufacturing, and Applications* (pp. 80–94). Wiley Blackwell. https://doi.org/10.1002/9781119248491.ch4

Guajardo, N., Grez, P., Schrebler, R. S., & Schrebler, R. A. (2025). Applications of Fluidized Bed Reactors in Biocatalysis. *ChemBioChem*, *26*(18). https://doi.org/10.1002/cbic.202500143

Guo, H., Zhao, Y., Chang, J. S., & Lee, D. J. (2022). Inhibitor formation and detoxification during lignocellulose biorefinery: A review. *Bioresource Technology*, *361*, 127666. https://doi.org/10.1016/J.BIORTECH.2022.127666

Han, W., Liu, Y., Xu, X., Huang, J., He, H., Chen, L., Qiu, S., Tang, J., & Hou, P. (2020). Bioethanol production from waste hamburger by enzymatic hydrolysis and fermentation. *Journal of Cleaner Production*, *264*. https://doi.org/10.1016/j.jclepro.2020.121658

Hermiati, E. (Euis), Mangunwidjaja, D. (Djumali), Sunarti, T. C. (Titi), Suparno, O. (Ono), & Prasetya, B. (Bambang). (2010). Pemanfaatan Biomassa Lignoselulosa Ampas Tebu Untuk Produksi Bioetanol. *Jurnal Penelitian Dan Pengembangan Pertanian*, *29*(4), 121–130. https://www.neliti.com/publications/178805/

Horváthová, M., Oravec, J., & Bakošová, M. (2020). Efficient convex-lifting-based robust control of a chemical reactor. *Chemical Engineering Transactions*, *81*, 865–8670. https://doi.org/10.3303/CET2081145

Hossain, M. T. (2023). Synthetic biology and metabolic engineering for improvement of lactic acid bacteria as cell factories. In *Lactic Acid Bacteria as Cell Factories: Synthetic Biology and Metabolic Engineering* (pp. 17–28). Elsevier. https://doi.org/10.1016/B978-0-323-91930-2.00006-7

Huang, H., Guo, X., Li, D., Liu, M., Wu, J., & Ren, H. (2011). Identification of crucial yeast inhibitors in bio-ethanol and improvement of fermentation at high pH and high total solids. *Bioresource Technology*, *102*(16), 7486–7493. https://doi.org/10.1016/j.biortech.2011.05.008

Huang, W. C., & Tang, I. C. (2006). Bacterial and Yeast Cultures-Process Characteristics, Products, and Applications. In *Bioprocessing for Value-Added Products from Renewable Resources: New Technologies and Applications* (pp. 185–223). Elsevier. https://doi.org/10.1016/B978-044452114-9/50009-8

Humera Farheen, V., Aslam Abdullah, M., & Ganesh Moorthy, I. (2026). Lignocellulosic bioethanol production: a review on pretreatment strategies, biofuel separation, and artificial intelligence/machine learning – based sustainable optimization. *Current Research in Biotechnology*, *11*. https://doi.org/10.1016/j.crbiot.2025.100355

Imamoglu, E., & Sukan, F. V. (2013). Scale-up and kinetic modeling for bioethanol production. *Bioresource Technology*, *144*, 311–320. https://doi.org/10.1016/j.biortech.2013.06.118

Iram, M., Asghar, U., Irfan, M., Huma, Z., Jamil, S., Nadeem, M., & Syed, Q. (2018). Production of bioethanol from sugarcane bagasse using yeast strains: A kinetic study. *Energy Sources, Part A: Recovery, Utilization and Environmental Effects*, *40*(3), 364–372. https://doi.org/10.1080/15567036.2017.1422056

Iriel, K. R. I., Mizan, M. G. P., & M. Iqbal Samudra. (2024). Microwave-Assisted Hydrolysis of Robusta Coffee Skin Waste as a Reducing Sugar Feedstock. *Journal of Biobased Chemicals*, *4*(1), 48–68. https://doi.org/10.19184/JOBC.V4I1.818

Iyabode Bright, A., & Olayinka, O. H. (2020). Production of Bioethanol From Groundnut (Arachis Hypogeae L.) Peels and Shells. *International Journal of Engineering Applied Sciences and Technology*, *04*(11), 38–44. https://doi.org/10.33564/ijeast.2020.v04i11.008

Jacobson, T. B., Adamczyk, P. A., Stevenson, D. M., Regner, M., Ralph, J., Reed, J. L., & Amador-Noguez, D. (2019). 2H and 13C metabolic flux analysis elucidates in vivo thermodynamics of the ED pathway in Zymomonas mobilis. *Metabolic Engineering*, *54*, 301–316. https://doi.org/10.1016/j.ymben.2019.05.006

Jalilnejad, E., Sadeghpour, P., & Ghasemzadeh, K. (2019). Advances in membrane bioreactor technology. In *Current Trends and Future Developments on (Bio-) Membranes: Ceramic Membrane Bioreactors* (pp. 1–29). Elsevier. https://doi.org/10.1016/B978-0-12-816822-6.00001-X

Jang, N., Yasin, M., Lee, M., Kang, H., & Chang, I. S. (2020). Gas circulation rate and medium exchange ratio as influential factors affecting ethanol production in carbon monoxide fermentation using a packed-bed reactor. *Sustainable Energy and Fuels*, *4*(4), 1963–1973. https://doi.org/10.1039/c9se00943d

Jeevitha, P., Ranjitha, J., Anand, M., Mahboob, S., & Vijayalakshmi, S. (2023). Production of pyruvic acid into value-added products using genetically modified microbes. In *Valorization of Biomass to Bioproducts: Organic Acids and Biofuels* (pp. 117–134). Elsevier. https://doi.org/10.1016/B978-0-12-822888-3.00012-8

Jefferson, B., McAdam, E., & Pidou, M. (2018). Membrane-based processes. In *Advances in Wastewater Treatment* (pp. 205–230). IWA Publishing. https://doi.org/10.2166/9781780409719_0205

Jia, X., Liu, D., Lin, H., Zhang, H., Liang, X., Ding, K., Ji, G., Han, L., & Xiao, W. (2025). Enhancing efficiency in ethanol production from high solid corn stover: Insights into enzymatic hydrolysis parameters and cellulase recovery. *Journal of the Taiwan Institute of Chemical Engineers*, *177*. https://doi.org/10.1016/j.jtice.2024.105644

John, A. J., & Selvarajan, E. (2024). Ionic liquid-assisted pretreatment of lignocellulosic biomass using purified Streptomyces MS2A cellulase for bioethanol production. *International Journal of Biological Macromolecules*, *270*(P1), 132149. https://doi.org/10.1016/j.ijbiomac.2024.132149

John J, A., Selvarajan, E., Kumar, M., & Samuel, M. S. (2026). Enhancing bioethanol production through hydrolysis with Streptomyces MS2A cellulase immobilized in the MIL-96AL-Fe3O4-GO-CS nanohybrid. *Biomass and Bioenergy*, *204*(September 2025), 108425. https://doi.org/10.1016/j.biombioe.2025.108425

Jugwanth, Y., Sewsynker-Sukai, Y., & Gueguim Kana, E. B. (2020). Valorization of sugarcane bagasse for bioethanol production through simultaneous saccharification and fermentation: Optimization and kinetic studies. *Fuel*, *262*(August 2019), 116552. https://doi.org/10.1016/j.fuel.2019.116552

Kadic, E., & Heindel, T. J. (2010). Mixing considerations in stirred tank bioreactors when using fluid property altering microorganisms. *American Society of Mechanical Engineers, Fluids Engineering Division (Publication) FEDSM*, *1*(PARTS A, B AND C), 859–870. https://doi.org/10.1115/FEDSM-ICNMM2010-30366

Kaewchana, A., Techaparin, A., Boonchot, N., Thanonkeo, P., & Klanrit, P. (2021). Improved high-temperature ethanol production from sweet sorghum juice using Zymomonas mobilis overexpressing groESL genes. *Applied Microbiology and Biotechnology*, *105*(24), 9419–9431. https://doi.org/10.1007/s00253-021-11686-0

Kamande, S. M., Omwenga, G. I., & Ngugi, M. P. (2024). Production of cellulases by Xylaria sp. and Nemania sp. using lignocellulose substrates for bioethanol production from maize cobs. *Heliyon*, *10*(17), e36802. https://doi.org/10.1016/j.heliyon.2024.e36802

Kassim, M. A., Meng, T. K., Kamaludin, R., Hussain, A. H., & Bukhari, N. A. (2021). Bioprocessing of sustainable renewable biomass for bioethanol production. In *Value-Chain of Biofuels: Fundamentals, Technology, and Standardization* (pp. 195–234). Elsevier. https://doi.org/10.1016/B978-0-12-824388-6.00004-X

Kawada-Matsuo, M., Oogai, Y., & Komatsuzawa, H. (2017). Sugar allocation to metabolic pathways is tightly regulated and affects the virulence of Streptococcus mutans. *Genes*, *8*(1). https://doi.org/10.3390/genes8010011

Keerthi Patel M, Muthuraju, R., Raj, S. M. N., Murali, M. K., Gowda, N. S., & Ashwini. (2025). Simultaneous saccharification and fermentation using lignocellulolytic microorganisms and Zymomonas mobilis for bioethanol production from paddy straw. *Biomass Conversion and Biorefinery*, *15*(16), 23117–23125. https://doi.org/10.1007/s13399-025-06764-6

Kobayashi, Y., Sahara, T., Ohgiya, S., Kamagata, Y., & Fujimori, K. E. (2018). Systematic optimization of gene expression of pentose phosphate pathway enhances ethanol production from a glucose/xylose mixed medium in a recombinant Saccharomyces cerevisiae. *AMB Express*, *8*(1). https://doi.org/10.1186/s13568-018-0670-8

Kreetachat, T., Suriyachai, N., Khongchamnan, P., Suwannahong, K., Wongcharee, S., Sakulthaew, C., Chokejaroenrat, C., & Imman, S. (2025). Optimization of Acid-Catalyzed Hydrolysis and Simultaneous Saccharification and Fermentation for Enhanced Ethanol Production from Sweet Stalk Sorghum. *Catalysts*, *15*(4), 1–17. https://doi.org/10.3390/catal15040379

Kumar, P., Kumar, V., Kumar, S., Singh, J., & Kumar, P. (2020). Bioethanol production from sesame (Sesamum indicum L.) plant residue by combined physical, microbial and chemical pretreatments. *Bioresource Technology*, *297*(November 2019), 122484. https://doi.org/10.1016/j.biortech.2019.122484

Li, J., Li, P., Xu, Y., & Wu, W. (2023). Mechanism and Research Progress of Lignin Affecting the Enzymatic Hydrolysis of Lignocellulosic Raw Materials[木质素影响木质纤维原料酶解的作用机理及其研究进展]. *Chung-Kuo Tsao Chih/China Pulp and Paper*, *42*(3), 114–120. https://doi.org/10.11980/j.issn.0254-508X.2023.03.015

Li, J., Shi, S., Tu, M., Via, B., Sun, F. F., & Adhikari, S. (2018). Detoxification of Organosolv-Pretreated Pine Prehydrolysates with Anion Resin and Cysteine for Butanol Fermentation. *Applied Biochemistry and Biotechnology*, *186*(3), 662–680. https://doi.org/10.1007/s12010-018-2769-4

Li, Y. F., Shu, Z., Chen, H., & Di, X. Y. (2011). The operation characteristics of biohydrogen production in continuous stirred tank reactor with molasses. *Advanced Materials Research*, *152–153*, 613–618. https://doi.org/10.4028/www.scientific.net/AMR.152-153.613

Li, Z., Lu, H., Zhang, Z., & Liu, B. (2023). Study on Scale-Up of Anaerobic Fermentation Mixing with Different Solid Content. *Fermentation*, *9*(4). https://doi.org/10.3390/fermentation9040375

Liguori, R., & Faraco, V. (2016). Biological processes for advancing lignocellulosic waste biorefinery by advocating circular economy. *Bioresource Technology*, *215*, 13–20. https://doi.org/10.1016/J.BIORTECH.2016.04.054

Lis, S., Tomasik, M., Szul, T., & Łyszczarz, P. (2020). The analysis of the automatic control of the bioethanol production process[Metodyka optymalizacji algorytmu sterowania w aspekcie ograniczenia zużycia energii w procesie wytwarzania bioetanolu]. *Przeglad Elektrotechniczny*, *96*(1), 214–217. https://doi.org/10.15199/48.2020.01.48

Liu, Q., Zhao, N., Zou, Y., Ying, H., Liu, D., & Chen, Y. (2020). Feasibility study on long-term continuous ethanol production from cassava supernatant by immobilized yeast cells in packed bed reactor. *Journal of Microbiology and Biotechnology*, *30*(8), 1227–1234. https://doi.org/10.4014/jmb.1908.08017

Liu, Y., Lv, M., Chen, R., Zhang, Z., Teng, R., Qin, J., Sun, M., Fan, Z., & Du, J. (2024). A peptide-based conductive hydrogel capable of in situ bioelectrocatalytic NADH regeneration for sustained production of 1-propanol. *Journal of Materials Chemistry A*, *13*(4), 2780–2788. https://doi.org/10.1039/d4ta07027e

Liu, Z., Fels, M., Dragone, G., & Mussatto, S. I. (2021). Effects of inhibitory compounds derived from lignocellulosic biomass on the growth of the wild-type and evolved oleaginous yeast Rhodosporidium toruloides. *Industrial Crops and Products*, *170*. https://doi.org/10.1016/j.indcrop.2021.113799

Liu, Z. L., Saha, B. C., & Slininger, P. J. (2014). Lignocellulosic Biomass Conversion to Ethanol by Saccharomyces. In *Bioenergy* (pp. 17–36). wiley. https://doi.org/10.1128/9781555815547.ch2

López-Caamal, F., Cea-Barcia, G., Hernández-Escoto, H., & Torres-Zúñiga, I. (2024). Optimizing Bioethanol Production via Extremum Seeking Control in a Continuous Stirred Tank Bioreactor. In *Springer Water: Part F3093* (pp. 187–215). Springer Nature. https://doi.org/10.1007/978-3-031-57735-2_10

Lorenci Woiciechowski, A., Dalmas Neto, C. J., Porto de Souza Vandenberghe, L., de Carvalho Neto, D. P., Novak Sydney, A. C., Letti, L. A. J., Karp, S. G., Zevallos Torres, L. A., & Soccol, C. R. (2020). Lignocellulosic biomass: Acid and alkaline pretreatments and their effects on biomass recalcitrance – Conventional processing and recent advances. *Bioresource Technology*, *304*. https://doi.org/10.1016/j.biortech.2020.122848

Lou, J., Wang, X., Li, R., Yao, J., Hu, J., Qin, X., Jin, M., & Yang, S. (2025). Integration of metabolic and evolutionary processes to construct efficient xylose-utilizing strain of Zymomonas mobilis for lignocellulosic ethanol production. *Synthetic and Systems Biotechnology*, *10*(3), 858–867. https://doi.org/10.1016/j.synbio.2025.04.009

Machado, T. J., Diniz, A. A. R., & Santos, M. D. dos. (2025). Artificial Intelligence and Machine Learning in Biofuels as Tools for Advancing Efficiency and Sustainability. In *Artificial Intelligence for Biomass-based Biofuel Production: Current Status and Prospects* (pp. 278–300). CRC Press. https://doi.org/10.1201/9781003571223-12

Maheshwari, N. V. (2018). Agro-industrial lignocellulosic waste: An alternative to unravel the future bioenergy. In *Biofuels: Greenhouse Gas Mitigation and Global Warming: Next Generation Biofuels and Role of Biotechnology* (pp. 291–305). Springer India. https://doi.org/10.1007/978-81-322-3763-1_16

Maibam, P. D., & Goyal, A. (2025). Bioethanol production from delignified rice straw by using a novel crude recombinant enzyme cocktail in pre-saccharification simultaneous saccharification and fermentation process. *Process Biochemistry*, *153*(December 2024), 26–33. https://doi.org/10.1016/j.procbio.2025.03.005

Makki, D. S., Majdi, H. S., Abdulrahman, A. A., Sultan, A. J., Hasan, Z. W., Sabri, L. S., Kadhim, B. J., & Al-Dahhan, M. H. (2023). A Comprehensive Review of the Influence of Heat Exchange Tubes on Hydrodynamic, Heat, and Mass Transfer in Bubble and Slurry Bubble Columns. *Fluid Dynamics and Materials Processing*, *19*(10), 2613–2637. https://doi.org/10.32604/fdmp.2023.028081

Malik, K., Salama, E. S., El-Dalatony, M. M., Jalalah, M., Harraz, F. A., Al-Assiri, M. S., Zheng, Y., Sharma, P., & Li, X. (2021). Co-fermentation of immobilized yeasts boosted bioethanol production

from pretreated cotton stalk lignocellulosic biomass: Long-term investigation. *Industrial Crops and Products*, *159*(November 2020), 113122. https://doi.org/10.1016/j.indcrop.2020.113122

Manmai, N., Unpaprom, Y., Ponnusamy, V. K., & Ramaraj, R. (2020). Bioethanol production from the comparison between optimization of sorghum stalk and sugarcane leaf for sugar production by chemical pretreatment and enzymatic degradation. *Fuel*, *278*(March), 118262. https://doi.org/10.1016/j.fuel.2020.118262

Mardawati, E., Putri, A. V., Yuliana, T., Rahimah, S., Nurjanah, S., & Hanidah, I. (2019). Effects of substrate concentration on bioethanol production from oil palm empty fruit bunches with simultaneous saccharification and fermentation (SSF). *IOP Conference Series: Earth and Environmental Science*, *230*(1). https://doi.org/10.1088/1755-1315/230/1/012079

Martín, M., Sánchez, A., & Woodley, J. M. (2018). Fermentative alcohol production. In *Green Energy and Technology* (Vol. 0, Number 9789811076763, pp. 319–357). Springer Verlag. https://doi.org/10.1007/978-981-10-7677-0_8

Mitra, R., Bhattacharya, I., Bhar, R., Dubey, B. K., Ghosh, A., & Sen, R. (2025). Strategic conversion of algal and lignocellulosic biomass wastes as mixed feedstock for enhanced and environment-friendly production of bioethanol. *Energy Conversion and Management*, *342*(June). https://doi.org/10.1016/j.enconman.2025.120157

Mohd Azhar, S. H., Abdulla, R., Jambo, S. A., Marbawi, H., Gansau, J. A., Mohd Faik, A. A., & Rodrigues, K. F. (2017). Yeasts in sustainable bioethanol production: A review. *Biochemistry and Biophysics Reports*, *10*, 52–61. https://doi.org/10.1016/J.BBREP.2017.03.003

Morales-Vera, R., Vásquez-Ibarra, L., Scott, F., Puettmann, M., & Gustafson, R. (2022). Life Cycle Assessment of Bioethanol Production: A Case Study from Poplar Biomass Growth in the U.S. Pacific Northwest. *Fermentation*, *8*(12), 734. https://doi.org/10.3390/FERMENTATION8120734/S1

Ningthoujam, R., Jangid, P., Yadav, V. K., Sahoo, D. K., Patel, A., & Dhingra, H. K. (2023). Bioethanol production from alkali-pretreated rice straw: effects on fermentation yield, structural characterization, and ethanol analysis. *Frontiers in Bioengineering and Biotechnology*, *11*(August), 1–9. https://doi.org/10.3389/fbioe.2023.1243856

Nishida, V. S., Woiciechowski, A. L., Valladares-Diestra, K. K., Zevallos Torres, L. A., Vandenberghe, L. P. de S., Zandoná Filho, A., & Soccol, C. R. (2023). Second Generation Bioethanol Production from Soybean Hulls Pretreated with Imidazole as a New Solvent. *Fermentation*, *9*(2). https://doi.org/10.3390/fermentation9020093

Norhazimah, A. H., Noh, T. U., & Mohd Noor, S. F. (2025). Optimization of fermentation conditions for bioethanol production from oil palm trunk sap. *Journal of the Indian Chemical Society*, *102*(9), 101943. https://doi.org/10.1016/j.jics.2025.101943

Nurcholis, M., Lertwattanasakul, N., Rodrussamee, N., Kosaka, T., Murata, M., & Yamada, M. (2020). Integration of comprehensive data and biotechnological tools for industrial applications of Kluyveromyces marxianus. *Applied Microbiology and Biotechnology*, *104*(2), 475–488. https://doi.org/10.1007/s00253-019-10224-3

Nury, D. F., Luthfi, M. Z., & Widjaja, T. (2024). PENGARUH KOMBINASI PRETREATMENT, HIDROLISIS, DAN FERMENTASI TERHADAP PRODUKSI BIOETANOL DARI LIMBAH PADAT AREN. *JURNAL INTEGRASI PROSES*, *13*(2), 201–207. https://doi.org/10.62870/JIP.V13I2.28466

Nwaze, I. O., Emmanuel, A., Nwaze, I. O., & Akubueze, E. U. (2023). Suitability of Groundnut Shells for Bioethanol Production using Saccharomyces cerevisae. *Journal of Materials Science Research and Reviews*, *11*(2), 34–39.

Ochoa-Chacón, A., Martinez, A., Poggi-Varaldo, H. M., Villa-Tanaca, L., Ramos-Valdivia, A. C., & Ponce-Noyola, T. (2022). Xylose Metabolism in Bioethanol Production: Saccharomyces cerevisiae vs Non-Saccharomyces Yeasts. *Bioenergy Research*, *15*(2), 905–923. https://doi.org/10.1007/s12155-021-10340-x

Padhan, B., Patel, G., Saha, P. S., Parween, I., & Das, J. (2025). High throughput microfluidic platforms for adaptive laboratory evolution of probiotic strains. In *Advances in Probiotic Delivery Systems: Strategies for Enhanced Viability, Targeted Delivery and Efficacy* (pp. 517–544). Elsevier. https://doi.org/10.1016/B978-0-443-27594-4.00003-6

Palupi, B., Aji, B. B., Prasiefa, M. G., Putri, D. K. Y., Rahmawati, I., Fachri, B. A., Rizkiana, M. F., & Amini, H. W. (2023). Microwave-Assisted Hydrolysis Batang Tembakau untuk Produksi Gula Pereduksi sebagai Bahan Baku Bioetanol. *Jurnal Teknologi*, *10*(2). https://doi.org/10.31479/JTEK.V10I2.225

Papathoti, N. K., Mendam, K., Thepbandit, W., Burgula, N., Sangpueak, R., Saengchan, C., Hoang, N. H., Keshav, P. K., Le Thanh, T., & Buensanteai, N. (2024). Bioethanol production from alkali-pretreated cassava stem waste via consolidated bioprocessing by ethanol-tolerant Clostridium thermocellum ATCC 31,924. *Biomass Conversion and Biorefinery*, *14*(5), 6821–6833. https://doi.org/10.1007/s13399-022-02868-5

Parawira, W., & Tekere, M. (2011). Biotechnological strategies to overcome inhibitors in lignocellulose hydrolysates for ethanol production: Review. *Critical Reviews in Biotechnology*, *31*(1), 20–31. https://doi.org/10.3109/07388551003757816

Peng, J., Xu, H., Wang, W., Kong, Y., Su, Z., & Li, B. (2021). Techno-economic analysis of bioethanol preparation process via deep eutectic solvent pretreatment. *Industrial Crops and Products*, *172*, 114036. https://doi.org/10.1016/J.INDCROP.2021.114036

Pino, M. S., Rodríguez-Jasso, R. M., Michelin, M., Flores-Gallegos, A. C., Morales-Rodriguez, R., Teixeira, J. A., & Ruiz, H. A. (2018). Bioreactor design for enzymatic hydrolysis of biomass under the biorefinery concept. *Chemical Engineering Journal*, *347*, 119–136. https://doi.org/10.1016/j.cej.2018.04.057

Qiang, Y., Wang, X., Liu, Y., Mao, H., Yang, L., Lv, Z., He, B., & Zhu, X. (2025). Construction of heterogeneous thermal biocatalytic system for high-performance cellulosic ethanol production. *International Journal of Biological Macromolecules*, *313*. https://doi.org/10.1016/j.ijbiomac.2025.144115

Quintero, J. A., Moncada, J., & Cardona, C. A. (2013). Techno-economic analysis of bioethanol production from lignocellulosic residues in Colombia: A process simulation approach. *Bioresource Technology*, *139*, 300–307. https://doi.org/10.1016/J.BIORTECH.2013.04.048

Rajesh Banu, J., Preethi, Kavitha, S., Tyagi, V. K., Gunasekaran, M., Karthikeyan, O. P., & Kumar, G. (2021). Lignocellulosic biomass based biorefinery: A successful platform towards circular bioeconomy. *Fuel*, *302*, 121086. https://doi.org/10.1016/J.FUEL.2021.121086

Rivera, E. C., De Farias, F., Atala, D. I. P., De Andrade, R. R., Da Costa, A. C., & Maciel, R. (2009). Development and implementation of an automated monitoring system for improved bioethanol production. *Chemical Engineering Transactions*, *18*, 451–456. https://doi.org/10.3303/CET0918073

Rodrigues, R. C. L. B., Canettieri, E. V., Prata, A. M. R., Martinez, E. A., José, Á. H. M., Palladino, F., da Silva, S. P., & Pessoa, A. (2024). Analytical Tools and Operational Requirements for Bioreactors. In *Bioreactor Technology in Food Processing* (pp. 59–83). CRC Press. https://doi.org/10.1201/9780429424236-4

Romaní, A., Tomaz, P. D., Garrote, G., Teixeira, J. A., & Domingues, L. (2016). Combined alkali and hydrothermal pretreatments for oat straw valorization within a biorefinery concept. *Bioresource Technology*, *220*(2016), 323–332. https://doi.org/10.1016/j.biortech.2016.08.077

S.K. Sharma, K.L. Kalra, and H. S. G. (2002). Fermentation of enzymatically saccharified sunflower stalks for ethanol production and its scale up. *Bioresource Technology*, *85*, 31–33. https://doi.org/https://doi.org/10.1016/S0960-8524(02)00076-7.

Saetang, N., & Tipnee, S. (2022). Influence of nanoparticles inclusion on the production of bioethanol from corn stalks and leaves. *Maejo International Journal of Energy and Environmental Communication*, *4*(2), 23–28. https://doi.org/10.54279/mijeec.v4i2.248138

Sarkar, N., Ghosh, S. K., Bannerjee, S., & Aikat, K. (2012). Bioethanol production from agricultural wastes: An overview. *Renewable Energy*, *37*(1), 19–27. https://doi.org/10.1016/J.RENENE.2011.06.045

Satwika, D., Djojoatmodjo, S., Finnegan, G., Puttajaya, D., & Ivana, J. (2019). Isolation and Characterization of Fermenting Yeast from Traditional Ethanol Production. *Journal of Physics: Conference Series*, *1397*(1). https://doi.org/10.1088/1742-6596/1397/1/012046

Sawhney, D., Vaid, S., Bangotra, R., Sharma, S., Dutt, H. C., Kapoor, N., Mahajan, R., & Bajaj, B. K. (2023). Proficient bioconversion of rice straw biomass to bioethanol using a novel combinatorial pretreatment approach based on deep eutectic solvent, microwave irradiation and laccase. *Bioresource Technology*, *375*(February), 128791. https://doi.org/10.1016/j.biortech.2023.128791

Scully, S. M., & Orlygsson, J. (2015). Recent advances in second generation ethanol production by thermophilic bacteria. *Energies*, *8*(1). https://doi.org/10.3390/en8010001

Sen, P., Nath, A., & Bhattacharjee, C. (2017). Packed-Bed Bioreactor and Its Application in Dairy, Food, and Beverage Industry. *Current Developments in Biotechnology and Bioengineering: Bioprocesses, Bioreactors and Controls*, 235–277. https://doi.org/10.1016/B978-0-444-63663-8.00009-4

Sharma, I. K. and A. D. (n.d.). *Bioreactor: Design, Functions and Fermentation innovations*. https://doi.org/10.5281/ZENODO.5775455

Sharma, L., Satya, P., Dev, B., Goswami, T., Alam, N. M., Roy, S., Singh, A. K., Pandey, S. K., Majumdar, B., Lavanya, A. K., & Kar, G. (2025). Enhanced bioethanol production from sequential alkali-laccase-hydroxybenzotriazole pretreatment and additive-mediated saccharification in kenaf (Hibiscus cannabinus L.). *Biomass and Bioenergy*, *201*(June), 108123. https://doi.org/10.1016/j.biombioe.2025.108123

Sharma, S., & Arora, A. (2020). Tracking strategic developments for conferring xylose utilization/fermentation by Saccharomyces cerevisiae. *Annals of Microbiology*, *70*(1). https://doi.org/10.1186/s13213-020-01590-9

Singh, A., & Kim, B. S. (2025). Nanoparticle-Assisted Bioethanol Production From Various Lignocellulosic Biomass. *International Journal of Energy Research*, *2025*(1). https://doi.org/10.1155/er/1937931

Singh, P., Gangwar, P., Kumar, N., & Ghosh, S. (2025). Continuous bioethanol production from glucose-rich hydrolysate derived from wheat straw using a unique fed-batch cultivation method in a bioreactor. *Process Safety and Environmental Protection*, *193*(July 2024), 74–86. https://doi.org/10.1016/j.psep.2024.11.047

Singh, P., Kiran, U., Dutta, B. C., Bhutani, S., & Ghosh, S. (2024). Bioconversion of hemicellulosic fraction of wheat straw biomass to bioethanol by Scheffersomyces stipitis: A kLa-based scale-up study. *Industrial Crops and Products*, *214*(January), 118461. https://doi.org/10.1016/j.indcrop.2024.118461

Soares, L. B., Bonan, C. I. D. G., Biazi, L. E., Dionísio, S. R., Bonatelli, M. L., Andrade, A. L. D., Renzano, E. C., Costa, A. C., & Ienczak, J. L. (2020). Investigation of hemicellulosic hydrolysate inhibitor resistance and fermentation strategies to overcome inhibition in non-saccharomyces species. *Biomass and Bioenergy*, *137*. https://doi.org/10.1016/j.biombioe.2020.105549

Sokan-Adeaga, A. A., Salami, S. A., Bolade, D. O., Aledeh, M., Sokan-Adeaga, M. A., Amubieya, O. E., Kehinde, S. A., Farzadkia, M., Ashraf, G. M., & Hoseinzadeh, E. (2024). Utilization of local corn (Zea Mays) wastes for bioethanol production by separate hydrolysis and fermentation. *Journal of Hazardous Materials Advances*, *15*(June). https://doi.org/10.1016/j.hazadv.2024.100447

Song, Y., Maskey, S., Lee, Y. G., Lee, D. S., Nguyen, D. T., & Bae, H. J. (2024). Optimizing bioconversion processes of rice husk into value-added products: D-psicose, bioethanol, and lactic acid. *Bioresource Technology*, *395*(November 2023), 130363. https://doi.org/10.1016/j.biortech.2024.130363

Srimachai, T., Meengam, C., Kongjan, P., & Rattanadilok Na Phuket, K. (2023). Efficient Conversion of Oil Palm Trunk and Frond to Bioethanol and Biogas Using Two-Stage Steam Explosion Pretreatment. *ASEAN Journal of Scientific and Technological Reports*, *26*(4), 11–20. https://doi.org/10.55164/ajstr.v26i4.249622

Srimachai, T., & Phuket, K. R. N. (2024). Influence of Inhibitors from Microwave Pretreatment of Oil Palm Frond Pulping (OPFP) on Bioethanol Production. *ASEAN Journal of Scientific and Technological Reports*, *27*(1), 1–14. https://doi.org/10.55164/ajstr.v27i1.249524

Sriramajayam, S., Padhi, T., Prabha, B., & Gitanjali, J. (2022). Bioethanol Production from Cotton Stalk through Simultaneous Saccharification and Fermentation System. *Chemical Science Review and Letters*, *11*(August), 325–333. https://doi.org/10.37273/chesci.cs205306487

Srivastava, A. (2025). Revolutionizing bioethanol production: The role of AI in process innovation. In *Methods in Microbiology* (Vol. 56, pp. 167–190). Academic Press Inc. https://doi.org/10.1016/bs.mim.2024.12.002

Straathof, A. J. J. (2023). Modelling of end-product inhibition in fermentation. *Biochemical Engineering Journal*, *191*. https://doi.org/10.1016/j.bej.2022.108796

Suhaimi, A. A., & Nasir, N. F. (2017). Study of the fluid flow pattern in a bubble column reactor for biodiesel production. *IOP Conference Series: Materials Science and Engineering*, *243*(1). https://doi.org/10.1088/1757-899X/243/1/012035

Sukhang, S., Choojit, S., Reungpeerakul, T. et al. (2020). Bioethanol production from oil palm empty fruit bunch with SSF and SHF processes using Kluyveromyces marxianus yeast. *Cellulose*, *27*, 301–314. https://doi.org/https://doi.org/10.1007/s10570-019-02778-2

Thangavelu, S. K. (2026). From response surface methodology to artificial intelligence: process intensification frameworks for sustainable bioethanol production. *Chemical Engineering and Processing - Process Intensification*, *221*, 110697. https://doi.org/10.1016/j.cep.2026.110697

Tutt, M., Kikas, T., & Olt, J. (2012). Influence of different pretreatment methods on bioethanol production from wheat straw. *Agronomy Research*, *10*(SPEC. ISS. 1), 269–276.

Tyupa, D. V., Morozov, A. N., Zakharov, Z. V., Kalenov, S. V., & Khamitov, R. A. (2021). Reconstruction of industrial processes for cultivating cho cells in a scale-down bioreactor model. *Biotekhnologiya*, *37*(4), 112–122. https://doi.org/10.21519/0234-2758-2021-37-4-112-122

Unyay, H., Perendeci, N. A., Piersa, P., Szufa, S., & Skwarczynska-Wojsa, A. (2024). Harnessing Switchgrass for Sustainable Energy: Bioethanol Production Processes and Pretreatment Technologies. *Energies*, *17*(19). https://doi.org/10.3390/en17194812

Vaithanomsat, P., Chuichulcherm, S., & Apiwatanapiwat, W. (2009). Bioethanol production from enzymatically saccharified sunflower stalks using steam explosion as pretreatment. *World Academy of Science, Engineering and Technology*, *37*(1), 140–143.

Valdivia-Rivera, S., Lizardi-Jiménez, M. A., Medina-Moreno, S. A., & Sánchez-Vázquez, V. (2019). Multiphase partitioning airlift bioreactors: An alternative for hydrocarbon biodegradation in contaminated environments. *Advances in Chemical Engineering*, *54*, 275–297. https://doi.org/10.1016/bs.ache.2019.01.006

Velvizhi, G., Balakumar, K., Shetti, N. P., Ahmad, E., Kishore Pant, K., & Aminabhavi, T. M. (2022). Integrated biorefinery processes for conversion of lignocellulosic biomass to value added materials: Paving a path towards circular economy. *Bioresource Technology*, *343*, 126151. https://doi.org/10.1016/J.BIORTECH.2021.126151

Venkatachalam, S. S. and S. (2025). An energy-efficient process for enhanced production of bioethanol from sorghum biomass: a futuristic approach towards circular economy. *Biomass Conv. Bioref*, *15*, 581–1592. https://doi.org/https://doi.org/10.1007/s13399-024-06083-2

Verardi, A., Blasi, A., Molino, A., Albo, L., & Calabrò, V. (2016). Improving the enzymatic hydrolysis of Saccharum officinarum L. bagasse by optimizing mixing in a stirred tank reactor: Quantitative analysis of biomass conversion. *Fuel Processing Technology*, *149*, 15–22. https://doi.org/10.1016/j.fuproc.2016.03.025

Verho, R., Londesborough, J., Penttilä, M., & Richard, P. (2003). Engineering Redox Cofactor Regeneration for Improved Pentose Fermentation in Saccharomyces cerevisiae. *Applied and Environmental Microbiology*, *69*(10), 5892–5897. https://doi.org/10.1128/AEM.69.10.5892-5897.2003

Wang, L. Q., Cai, L. Y., & Ma, Y. L. (2020). Study on inhibitors from acid pretreatment of corn stalk on ethanol fermentation by alcohol yeast. *RSC Advances*, *10*(63), 38409–38415. https://doi.org/10.1039/d0ra04965d

Wang, Q., & Cui, Z. (2025). Biotech Porous Membranes. In *Porous Membranes: Breakthroughs in Manufacturing and Applications* (pp. 175–206). wiley. https://doi.org/10.1002/9781394303489.ch7

Wang, X., Bai, X., Chen, D. F., Chen, F. Z., Li, B. Z., & Yuan, Y. J. (2015). Increasing proline and myo-inositol improves tolerance of Saccharomyces cerevisiae to the mixture of multiple lignocellulose-derived inhibitors. *Biotechnology for Biofuels*, *8*(1). https://doi.org/10.1186/s13068-015-0329-5

Xia, J., Yang, Y., Liu, C. G., Yang, S., & Bai, F. W. (2019). Engineering Zymomonas mobilis for Robust Cellulosic Ethanol Production. *Trends in Biotechnology*, *37*(9), 960–972. https://doi.org/10.1016/j.tibtech.2019.02.002

Xu, S., Liu, H., Li, X., Du, G., & Chen, J. (2022). Hydrodynamics and mass transfer in a three-phase bubble column cell culture bioreactor[鼓泡塔细胞反应器流体力学与传质特性]. *Guocheng Gongcheng Xuebao / The Chinese Journal of Process Engineering*, *22*(9), 1192–1202. https://doi.org/10.12034/j.issn.1009-606X.221364

Yanase, H. (2014). Ethanol from Bacteria. In *Bioprocessing of Renewable Resources to Commodity Bioproducts* (Vol. 9781118175835, pp. 167–199). Wiley Blackwell. https://doi.org/10.1002/9781118845394.ch7

Yang, S., Dong, X., & Wang, L. (2026). Techno-economic and life cycle assessment of a biobased solvent-driven lignocellulosic biomass refinery. *Journal of Cleaner Production*, *538*, 147321. https://doi.org/10.1016/J.JCLEPRO.2025.147321

Zapater, D., Kulkarni, S. R., Wery, F., Cui, M., Herguido, J., Menendez, M., Heynderickx, G. J., Van Geem, K. M., Gascon, J., & Castaño, P. (2024). Multifunctional fluidized bed reactors for process intensification. *Progress in Energy and Combustion Science*, *105*. https://doi.org/10.1016/j.pecs.2024.101176

Zeng, Y., Zhao, S., Yang, S., & Ding, S. Y. (2014). Lignin plays a negative role in the biochemical process for producing lignocellulosic biofuels. *Current Opinion in Biotechnology*, *27*, 38–45. https://doi.org/10.1016/J.COPBIO.2013.09.008

Zentou, H., Abidin, Z. Z., Zouanti, M., & Greetham, D. (2017). Effect of operating conditions on molasses fermentation for bioethanol production. *International Journal of Applied Engineering Research*. https://www.scopus.com/pages/publications/85028323002?origin=scopusAI

Zhang, Y., Xia, C., Lu, M., & Tu, M. (2018). Effect of overliming and activated carbon detoxification on inhibitors removal and butanol fermentation of poplar prehydrolysates. *Biotechnology for Biofuels*, *11*(1). https://doi.org/10.1186/s13068-018-1182-0

Zhang, Z., Jiang, L., Wang, H., Li, Q., Ali, S., Chen, Y., Tang, J., Shen, J., Xin, W., Feng, L., & Ma, M. (2026). Discovery of a lignocellulosic hydrolysate-tolerant Saccharomyces cerevisiae strain and elucidation of a novel stress resistance mechanism. *Biomass and Bioenergy*, *209*. https://doi.org/10.1016/j.biombioe.2026.108941

Zhao, J., Li, J., Dong, Z., & Shao, T. (2025). Study on various degradation mechanisms of lignocellulose during spontaneous ensiling and their contributions to 2G bioethanol production. *Industrial Crops and Products*, *229*(April), 121020. https://doi.org/10.1016/j.indcrop.2025.121020

Zhou, M., & Lü, X. (2021). Strategies on simultaneous fermentation of pentose and hexose to bioethanol. In *Advances in 2nd Generation of Bioethanol Production* (pp. 161–211). Elsevier. https://doi.org/10.1016/B978-0-12-818862-0.00010-8

Zhu, J., Chen, W., Chen, Y., Amin, F. R., Li, Y., Lu, M., & Li, D. (2025). CFD optimization of an air lift fermenter for Fusarium venenatum fermentation. *Bioresource Technology Reports*, *29*. https://doi.org/10.1016/j.biteb.2025.102024

Zhu, Q., Gao, D., Yan, D., Tang, J., Cheng, X., El Sayed, I. E. T., El-Gendy, N. S., Lu, X., & Xin, J. (2023). Highly efficient one-pot bioethanol production from corn stalk with biocompatible ionic

liquids. *Bioresource Technology Reports*, *22*(March), 101461. https://doi.org/10.1016/j.biteb.2023.101461