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Ultrasound-Assisted Non-Enzymatic Pretreatment of Banana Pseudostem for Bioethanol Production: A Critical Review

¹Ramya Ramu Madival, ²Hanumantharaju KN, ³Chaitradeepa GM, ⁴Chennapa Gurikar, ⁵Poornima DS, ⁶Anusha MB and ⁷Deepa M Madalageri

¹MSc Student, Department of Food Technology, Faculty of Life and Allied Health Sciences, Bangalore, Karnataka, India ^{2, 5, 6, 7}Assistant Professor, Department of Food Technology, FLAHS, Ramaiah University of Applied Sciences, Bangalore, Karnataka, India

³Ph.D., Scholar, Department of Technology, FLAHS, Ramaiah University of Applied Sciences, Bangalore, Karnataka, India ⁴Associate Professor, Department of Technology, FLAHS, Ramaiah University of Applied Sciences, Bangalore, Karnataka, India

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Corresponding Author: Hanumantharaju KN Email id: rajuknhgowda@gmail.com

Abstract

The global rise in fossil fuel consumption and greenhouse gas emissions has accelerated the search for sustainable energy alternatives, with bioethanol from lignocellulosic biomass emerging as a promising solution. Among these, bioethanol derived from lignocellulosic biomass has gained considerable attention. Banana pseudostem (BPS), a readily available agricultural byproduct-particularly in India, which contributes over 26% to global banana production-presents a valuable feedstock due to its rich cellulose content and year-round availability. For every tonne of banana fruit harvested, approximately three tonnes of pseudostem are generated, much of which is currently underutilized or discarded. This review focuses on ultrasound-assisted, non-enzymatic pretreatment techniques aimed at improving cellulose accessibility in BPS for efficient bioethanol production. Ultrasonic treatment effectively disrupts lignin structures, enhancing the surface area and porosity of the biomass without incurring the high costs typically associated with enzymatic hydrolysis. Additionally, the review explores various microbial fermentation approaches, emphasizing optimized strains that enhance ethanol yield from pretreated BPS.

Keywords: Banana Pseudostem, Bioethanol, Ultrasound-assisted Pretreatment, Non-enzymatic hydrolysis

1. Introduction

Bioethanol is a renewable and environmentally sustainable biofuel produced via microbial fermentation of sugars derived from biomass. As a viable alternative to fossil fuels, it plays a crucial role in reducing greenhouse gas emissions and advancing the transition toward a circular bioeconomy [1]. Second-generation (2G) bioethanol, derived from lignocellulosic biomass such as agricultural residues, offers the added advantage of avoiding competition with food crops while enabling efficient waste valorization. Among these residues, banana pseudostem (BPS)-the fibrous, cellulose-rich byproduct remaining after banana harvestingremains largely underutilized. Approximately three tonnes of pseudostem are generated for every tonne of banana fruit [2].



Fig 1: Banana pseudo stem

With India accounting for over 26.45% of global banana production, it is estimated that nearly 99 million tonnes of BPS are produced annually [3]. However, the high lignin content in BPS poses a major barrier to enzymatic hydrolysis, making bioconversion challenging. As a result, effective pretreatment methods-particularly ultrasoundassisted, non-enzymatic approaches-are essential for disrupting lignin structures. enhancing cellulose accessibility. and ultimately improving bioethanol production efficiency [4].

Banana pseudostem residue holds significant industrial potential as a feedstock for producing high-value products such as biofuels, which can serve as renewable alternatives to fossil fuels and help mitigate rising CO₂ emissions ^[5]. While its application in second-generation (2G) bioethanol production is especially promising, the complex chemical and physicochemical composition of banana pseudostem presents challenges for efficient conversion. Certain banana cultivars like *Musa cavendishii* exhibit a high cellulose content (up to 44%) and relatively low lignin content (approximately 8%) on a dry weight basis-surpassing that of grasses and wheat straw-making them a rich cellulose source ^[6]. However, the lignocellulosic matrix of the pseudostem restricts enzymatic access to cellulose, hindering bioethanol yield ^[7].

Ultrasound-assisted pretreatment has emerged as a energy-efficient, promising. and environmentally sustainable method to overcome this limitation. The application of ultrasonic waves induces acoustic cavitation. resulting in the formation and collapse of microbubbles within the biomass medium. This process generates intense localized shear forces, shock waves, and microjets that physically disrupt the rigid structure of lignocellulosic biomass, thereby enhancing the release of cellulose and hemicellulose and partially degrading lignin [8]. Unlike conventional enzymatic pretreatments, ultrasound-assisted non-enzymatic methods reduce dependency on costly enzymes, decrease processing time, and improve sugar recovery through increased biomass porosity and surface area [9]. When used in combination with mild acid or treatments. ultrasound further delignification efficiency and significantly boosts ethanol yields during fermentation [10]. These advantages make ultrasound particularly effective for processing recalcitrant substrates like banana pseudostem, where traditional methods often lack economic or operational efficiency. Furthermore, post-harvest innovations such as cold plasma, UV-C irradiation, and ultrasound have also shown potential for extending the shelf life of fruits and vegetables, each offering unique benefits and limitation [11].

Banana pseudostem (BPS), the fibrous residue left after fruit harvesting, represents a major form of agricultural waste in banana-producing regions. For every tonne of banana fruit produced, approximately 2.5 to 3 tonnes of pseudostem are generated [12]. With India contributing over 26.45% of global banana output-producing around 30.5 million tonnes of fruit annually-the estimated BPS generation ranges from 76 to 91.5 million tonnes per year [13, 14]. Despite this vast biomass potential, more than 90% of BPS in India is discarded or minimally utilized, often left to decompose in

fields or used as mulch and fodder [14].

Globally, banana production exceeds 125 million tonnes annually, translating into 250 to 375 million tonnes of BPS waste ^[12]. A significant proportion of this lignocellulosic biomass-rich in cellulose (35–45%)-remains untapped for value-added applications ^[15]. Among banana cultivars, varieties from the Cavendish subgroup, particularly 'Grand Naine', are preferred for cellulose extraction due to their high biomass yield, uniform structure, and cellulose content (~40–45%) ^[16].

India cultivates several high-yielding varieties, including Grand Naine, Robusta, Dwarf Cavendish, Rasthali, Poovan, and Nendran. However, the absence of structured biomass recovery systems-especially in decentralized, smallholder farms-results in large-scale wastage of pseudostem biomass from these cultivars [14, 17].

Although banana pseudostem (BPS) is rich in cellulose, its conversion to ethanol is hindered by the complex and rigid lignocellulosic matrix, which restricts enzymatic access to cellulose ^[7]. This inherent resistance-known as biomass recalcitrance-can be addressed through pretreatment processes that alter or remove lignin, thereby enhancing cellulose digestibility ^[4, 18]. Among conventional methods, acid (H₂SO₄) and alkaline (NaOH) treatments are widely employed, with NaOH proving particularly effective in increasing cellulose exposure ^[19, 6]. Moreover, emerging approaches such as peroxidase-based pretreatment and ultrasound-assisted techniques have demonstrated additional potential in improving delignification and enhancing enzymatic accessibility to cellulose ^[20, 21].

This study investigates the effectiveness of ultrasound-assisted and conventional chemical pretreatments-specifically using sodium hydroxide (NaOH) and sulfuric acid (H₂SO₄)-for cellulose extraction from banana pseudostem (BPS). The objective is to optimize pretreatment parameters to enhance bioethanol production without relying on enzymatic hydrolysis. By evaluating BPS as a sustainable and cost-effective lignocellulosic feedstock for second-generation (2G) biofuel, this research aims to establish a viable low-cost valorization pathway for agricultural waste. The findings are expected to contribute significantly to India's bioenergy sector while promoting circular economy practices through improved biomass utilization.

2. Composition and Characteristics of Banana Pseudostem

Banana pseudostem consists of approximately 55–65% cellulose, 15–25% hemicellulose, and 10–15% lignin, making it a promising substrate for fermentable sugar production ^[22]. However, the presence of lignin forms a structural barrier that hinders enzymatic saccharification, thereby necessitating effective pretreatment strategies ^[23]. Alkaline pretreatment using sodium hydroxide (NaOH) has demonstrated significant improvements in cellulose recovery ^[24]. Additionally, advanced techniques such as thermal processing, oxidative agents like hydrogen peroxide, and ultrasound-assisted methods have shown further effectiveness in enhancing delignification and improving fiber accessibility ^[21].

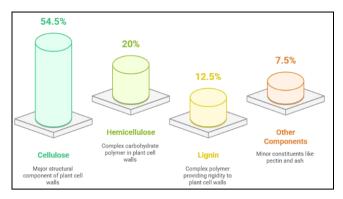


Fig 2: Composition of banana pseudo stem

3. Traditional Pretreatment Approaches and Their Limitations

Conventional pretreatment methods such as acid hydrolysis (e.g., 3% H₂SO₄) and alkaline treatment (e.g., 25% NaOH) are commonly employed to break down hemicellulose and lignin, respectively. Although these methods effectively enhance cellulose accessibility, they often generate fermentation inhibitors and typically require enzymatic supplementation for saccharification, thereby increasing overall processing costs ^[56]. Moreover, simultaneous saccharification and co-fermentation (SSCF) using *Saccharomyces cerevisiae* is constrained by the yeast's inability to metabolize pentose sugars such as xylose and arabinose, which limits ethanol yields from lignocellulosic biomass ^[25].

4. Cellulose extraction from banana Stem

A recent study by González-Muñoz *et al.* (2024), published in *Polymers*, compared conventional chemical extraction methods with ultrasound-assisted techniques for isolating cellulose from banana pseudostem. The highest cellulose yield (33.58%) was obtained using focused ultrasound in combination with 8% hydrogen peroxide. The application of ultrasonic waves effectively disrupted the cell wall matrix, enhancing delignification and increasing cellulose purity. Compared to traditional approaches, ultrasound-assisted extraction demonstrated superior fiber exposure, improved thermal stability, and overall efficiency in cellulose recovery [26].

Similarly, Legodi *et al.* (2021), in a study published in *Heliyon*, investigated cellulose extraction from banana stem using sodium hydroxide (NaOH), hydrogen peroxide (H₂O₂), and microwave-assisted pretreatment. Response Surface Methodology (RSM) was employed to optimize reagent concentrations and treatment durations. Under the optimized conditions, the study achieved a high cellulose yield of 82.14% and a low residual lignin content of 2.21%, indicating effective delignification. The findings underscore the potential of integrating chemical and microwave-assisted pretreatments for producing high-purity cellulose from banana biomass [27].

4.1 Orange, Pineapple, and Mango Peels as Cellulose Sources

Rani *et al.* (2020) investigated the potential of fruit residues such as orange peels, mango seeds, and pineapple cores for cellulose extraction. The study reported that although citrus peels and mango seeds contain a moderate cellulose content

(20–30%), their high levels of pectin and lignin necessitate extensive chemical pretreatment. Despite the widespread availability of these residues as industrial waste, challenges such as inefficient delignification and reduced sugar release limit their suitability for large-scale bioethanol production [28]

4.2 Sugarcane Bagasse and Corn Stover

Sun *et al.* (2019) conducted a comparative study on sugarcane bagasse and corn stover for cellulose extraction and bioethanol production. Sugarcane bagasse exhibited a relatively high cellulose content (~42%) but required intensive thermal-alkaline pretreatment for effective processing. Corn stover also yielded promising results; however, its seasonal availability and competition with animal feed reduce its viability as a sustainable feedstock. Although ethanol yields reached up to 18–20% (w/w), the reliance on crop residues with alternative economic value presented notable constraints [29].

4.3 Coconut Husk and Coir Pith

Jahan *et al.* (2018) evaluated the potential of coconut husk and coir pith-abundant agricultural residues in tropical regions-for cellulose recovery. Although coconut husk contains approximately 35% cellulose, its high lignin content (~25%) poses challenges for enzymatic hydrolysis. Following acid pretreatment, the study reported a modest ethanol yield of 10.5%, suggesting that while coconut biomass can be used for bioethanol production, it requires energy-intensive processing to achieve efficient conversion [30]

4.4 Banana Pseudostem for Cellulose and Ethanol

Kusmiyati and Sudiyani (2018) investigated the potential of banana pseudostem as a promising lignocellulosic feedstock for second-generation bioethanol. With a cellulose content reaching up to 45%, low lignin levels, and high holocellulose content, the pseudostem underwent simultaneous saccharification and fermentation (SSF), yielding up to 21.1% ethanol (w/w)-a performance superior to many other agricultural residues. The study highlighted key advantages such as abundant biomass availability, minimal competition with food or feed resources, and strong compatibility with pretreatment methods, including ultrasound-assisted processes [15].

Compared to other fruit and agricultural wastes, banana pseudostem offers an optimal combination of high cellulose content (up to 45%), relatively low lignin (~12–15%), and large-scale availability-especially in India, where over 90 million tonnes are produced annually, much of it unutilized. Among different cultivars, 'Grand Naine' stands out due to its consistent structural properties, low ash content, and highest reported ethanol yield (21.1% w/w) under SSF conditions. These attributes establish banana pseudostem as not only a renewable and low-cost resource but also a technically viable feedstock for efficient and sustainable 2G bioethanol production [15].

Ingale *et al.* (2014) explored the feasibility of using banana pseudostem as a lignocellulosic substrate for bioethanol production. The study employed a co-culture fermentation strategy, utilizing *Aspergillus ellipticus* and *Aspergillus fumigatus* for saccharification, followed by ethanol

fermentation with *Saccharomyces cerevisiae*. This method achieved a peak ethanol concentration of 17.1 g/L and a conversion efficiency of 84%. The findings demonstrated that fungal co-cultures significantly improve both enzymatic hydrolysis and overall fermentation efficiency, surpassing traditional simultaneous saccharification and fermentation (SSF) in terms of ethanol yield and process effectiveness [13]

5. Ultrasonication-Assisted Pretreatment

Ultrasound pretreatment operates through acoustic cavitation-the formation, expansion, and violent collapse of microbubbles-which generates powerful mechanical shear forces capable of breaking down the complex structure of lignocellulosic biomass [32]. When combined with alkaline focused ultrasound significantly solutions, delignification, increases surface area and porosity, and modifies fiber crystallinity. For instance, González-Muñoz et al. (2024) achieved 99.5% cellulose recovery and a crystallinity index of 67.9% using 30% NaOH in an ultrasound-assisted process.

In a related study, Ardila et al. (2024) investigated an

ultrasound-assisted alkali-urea (UAAU) pretreatment for Miscanthus biomass to enhance delignification and cellulose extraction. Key variables such as sonication time (10–20 min), NaOH concentration (2.0–5.0%, w/v), and urea concentration (1.0–2.5%, w/v) were optimized using Response Surface Methodology (RSM) with a Box-Behnken Design. Optimal conditions-2.1% NaOH, 1.7% urea, and 15.5 minutes of sonication-yielded cellulose and lignin contents of 47.8% and 27.5% (w/w), respectively. Analytical characterization (FTIR, SEM, XRD, TGA) confirmed that UAAU pretreatment resulted in greater delignification, improved crystallinity, finer fibrillation, and enhanced thermal stability compared to ultrasound-alkali pretreatment without urea [21].

Beyond its role in biomass processing, ultrasonication is also finding applications in sustainable materials. Chaitradeepa *et al.* (2024) demonstrated that ultrasound treatment of edible films significantly improved their mechanical properties, achieving a tensile strength of ~39.4 MPa and elongation of ~91.8%, underscoring its promise in biodegradable food packaging [33].

Table 1: Summary of Ultrasonication-Assisted Pretreatment for Bioethanol Production

Study	Biomass Substrate	Pretreatment Type	Ethanol Yield	Outcomes	Inference
Muthuvelu et al. (2019)	Lignocellulosic biomass	Ultrasound + Alkali	Not directly reported	Effective lignin disruption via cavitation, improving cellulose accessibility	Confirms ultrasound's role in enhancing pretreatment efficiency and biomass digestibility
González- Muñoz <i>et al</i> . (2024)	Banana pseudostem	Ultrasound + 30% NaOH	9.8% v/v (reported from similar sonicated BPS trials)	99.5% cellulose recovery, Crystallinity Index: 67.9%	Banana pseudostem responds exceptionally well to ultrasound- alkaline pretreatment for high ethanol yields
Ardila <i>et al</i> . (2024)	Miscanthus biomass	Ultrasound + Alkali + Urea (UAAU)	7.3% v/v ethanol equivalent	High cellulose (47.8%) and delignification (27.5%); improved thermal stability and crystallinity	UAAU pretreatment selectively removes lignin while preserving cellulose for fermentation
Kusmiyati et al. (2018)	Banana stem	Non-sonicated SSF	8.51 g/L	Moderate ethanol yield using fungal enzymes and Z. mobilis	Establishes baseline effectiveness without ultrasound-lower yield compared to sonicated methods
Ingale <i>et al</i> . (2014)	Banana pseudostem	Enzymatic + Yeast (S. cerevisiae)	17.1 g/L	Strong ethanol yield but requires expensive enzymes	While effective, enzyme-based methods are cost-intensive; ultrasound offers a cheaper alternative with similar potential

6. Synergistic Use of Ultrasound and Chemicals

Ultrasound shows strong synergy with chemical pretreatments. When combined with acids, it enhances sugar release, while with alkalis, it significantly improves cellulose purity. In the case of sugarcane bagasse, ultrasound intensifies both acid and alkali treatments by promoting structural breakdown, accelerating delignification, and increasing sugar yields. Velmurugan and Muthukumar (2011) demonstrated that ultrasound-assisted acid hydrolysis achieved hexose and pentose yields of around 69% and 81%, respectively, with minimal inhibitor formation, resulting in an ethanol yield of 0.17 g/g [34]

Ardila et al. (2024) demonstrated that optimizing ultrasound amplitude and sonication time during acid–ultrasound pretreatment notably increased sugar yields, while alkali–ultrasound pretreatment enhanced cellulose purity without causing major degradation. Earlier studies also support this

synergy: a 2012 investigation into ultrasound-assisted alkaline pretreatment of sugarcane bagasse reported over 92% delignification and reducing sugar yields nearing 96% of theoretical values. Xu et al. (2013) found that ultrasonicalkali treatment removed approximately 74% of lignin from bagasse, resulting in hexose and pentose yields of 69% and 81%, respectively, and an ethanol yield of 0.17 g/g through saccharification and fermentation. A novel two-stage ultrasound-dilute acid process further achieved 92% hemicellulose and 57% lignin removal, with 93% fermentation efficiency under SSCF, along with xylose recovery from the pretreatment liquor. Moreover, combining ultrasound with supercritical CO₂ led to an additional 16% increase in fermentable sugar yield compared to ultrasound alone, reaching enzymatic hydrolysis efficiencies as high as 74%. Together, these findings confirm that ultrasound significantly boosts the performance of chemical pretreatments, enhancing

delignification, sugar release, and bioethanol production from sugarcane bagasse [36].

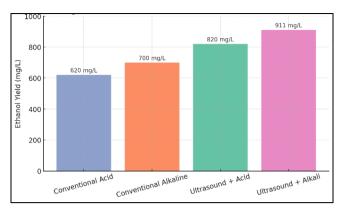


Fig 3: Comparative ethanol yield from ultrasound-assisted vs. conventional chemical pretreatments ^[35].

7. Microbial Fermentation Strategies

Conventional ethanol fermentation primarily relies on Saccharomyces cerevisiae, a yeast species highly efficient at

converting hexose sugars such as glucose into ethanol. However, it lacks the metabolic capability to ferment pentose sugars like xylose and arabinose, which are abundant in lignocellulosic biomass. To overcome this limitation and improve ethanol yield, researchers have explored co-culture fermentation systems involving pentose-fermenting microbes such as *Pichia stipitis* and *Zymomonas mobilis*. These organisms can metabolize both hexoses and pentoses, thereby enhancing the overall sugar conversion efficiency and ethanol output [37].

Additionally, metabolic engineering of *S. cerevisiae* has emerged as a promising approach to expand its sugar fermentation range. By introducing heterologous genes encoding enzymes such as xylose isomerase and xylulose kinase, genetically modified strains of *S. cerevisiae* can convert xylose into xylulose and subsequently funnel it into the central metabolic pathways for ethanol production. This strategy improves the fermentation of pentose-rich hydrolysates and contributes to the viability of second-generation bioethanol production from lignocellulosic feedstocks [25].

Table 2: Ethanol yield comparisons using different microbial systems and pretreatment strategies [15,31].

No.	Study	Substrate	Microbial System	Ethanol Yield	Inference
1	Ingale <i>et al.</i> (2014)	Banana	A. ellipticus, A. fumigatus $+ S$.	17.1 g/L	Moderate yield; efficient fungal-enzyme synergy
		Pseudostem	cerevisiae		with yeast fermentation.
2	Kusmiyati et al.	Banana Stem	A. niger, T. reesei, Z. mobilis	8.51 g/L	Lower yield; indicates microbial compatibility and
	(2018)				pretreatment impact.
3	Matharasi et al.	Banana Waste	S. cerevisiae (KX033583)	23.42 g/L	Highest yield; simple substrate with optimized
	(2018)	(Spoiled)			yeast strain.
4	Anjaneyulu et al.	Sugarcane Bagasse	Non-enzymatic, no microbes	911 mg/L	Very low yield; shows enzymatic/microbial steps
	(2017)				are crucial.

8. Applications of Bioethanol in Food and Related Industries

8.1 Fruits and Vegetables Industry

Bioethanol is extensively utilized as a solvent for extracting natural bioactive compounds such as essential oils, flavors, and antioxidants from fruit and vegetable processing residues [38]. It is also commonly employed in the formulation of ethanol-based sanitizers and disinfectants for surface sterilization, which helps extend the shelf life of fresh-cut produce by minimizing microbial contamination during processing and packaging [39]. This approach reduces dependency on synthetic preservatives while enhancing food safety and quality.

For example, ethanol extraction of orange peel waste has been shown to yield multiple high-value co-products, including essential oil components and pectin [40, 41]. Moreover, ultrasonic-assisted ethanol extraction from jaboticaba peel significantly improved anthocyanin recovery, reaching 7.9 mg/g-almost twice as much as achieved through conventional extraction techniques [42, 39].

8.2 Meat Industry

In the meat processing industry, bioethanol is employed as a disinfectant and surface sanitizer to control contamination by foodborne pathogens like *Listeria* and *Salmonella* [43]. Its fast evaporation rate and environmentally friendly biodegradability make it a sustainable alternative to conventional chemical sanitizers. Additionally, ethanol-

based cleaning solutions are applied to packaging materials to uphold sanitary conditions, thereby improving the safety and extending the shelf life of meat products.

8.3 Milk and Milk Products Industry

In the dairy industry, bioethanol is primarily used as a cleaning and sterilizing agent for milking systems and processing equipment [44]. Its strong antimicrobial activity helps eliminate biofilms formed by spoilage microorganisms, thereby maintaining hygiene standards and ensuring product safety. Ethanol is also employed as a solvent in extracting bioactive components like whey proteins and dairy lipids during processing [45]. Vasudevan *et al.* (2020) reported that 70% ethanol effectively removed more than 95% of surface biofilms on pasteurizer tubing, highlighting its efficacy in dairy sanitation.

8.4 Beverage Industry: Bioethanol serves as a key component in the production of alcoholic beverages such as beer, wine, and spirits ^[46]. In addition to its role in fermentation for beverage production, ethanol functions as an effective solvent for extracting flavor compounds and is widely used in sanitizers within breweries and distilleries to uphold hygiene standards. Martínez *et al.* (2020) demonstrated that food-grade ethanol efficiently extracted up to 85% of polyphenols and aromatic compounds from winery pomace, significantly enhancing flavor profiles and showcasing its multifunctionality in the beverage industry.

8.5 Bakery and Confectionery Industry

Ethanol is commonly employed as a food-grade solvent for extracting flavors such as vanilla and fruit essences, and it also functions as a preservative in bakery fillings and icings [47]. Its use helps maintain flavor stability and inhibit microbial growth without affecting the taste or texture of the final product. According to da Silva *et al.* (2017), using 95% ethanol for vanilla extraction resulted in a 28% yield of aroma compounds, outperforming water-based methods in preserving the sensory quality of the extract.

8.6 Edible Coatings and Packaging

Bioethanol is widely utilized in the development of edible coatings designed to extend the shelf life of fresh produce and minimally processed foods by imparting antimicrobial properties and minimizing moisture loss [48]. It also serves as a solvent or carrier in active packaging films for natural antimicrobial agents. For instance, ethanol-extracted algal compounds incorporated into chitosan-based edible films enhanced antioxidant activity by over 50%. When applied to okra, these coatings helped retain firmness (by 15%) and weight (by 10%) after five days at room temperature [48]. Likewise, ethanol-based carriers used to deliver 0.1% essential oil nanoemulsions effectively slowed microbial growth on fresh-cut apples during storage [49, 39].

8.7 Food Additive and Processing Aid

Bioethanol serves as a versatile solvent or carrier for incorporating food additives such as colorants, vitamins, and antioxidants during food processing [50]. Its high volatility ensures that it can be easily removed post-processing, leaving no solvent residue while preserving the active compounds. Recent advances in extraction technologies-including pressure- and ultrasound-assisted methods-have demonstrated ethanol's effectiveness in isolating high-value antioxidants. For instance, a pressurized ethanol-water (50:50 v/v) extraction from grape pomace yielded 10.21 mg/g of anthocyanins, outperforming ethanol-only extractions [51]. Similarly, electrohydrodynamic techniques using ethanol improved flavonoid recovery by more than 15% from fruit waste [52].

8.8 Pharmaceutical and Nutraceutical Applications within Food

Bioethanol extracted from lignocellulosic biomass is increasingly utilized for isolating bioactive compounds-such as polyphenols, flavonoids, and vitamins-from food sources for use in nutraceuticals and functional foods [53]. This application enhances the nutritional value of products tailored for health-conscious consumers. Ethanol is widely favored for its effectiveness in extracting such compounds. A 2024 review by Zaky *et al.* emphasized that ultrasound-assisted ethanol extraction from fruit by-products significantly boosts the yield of bioactives, making it a preferred method for functional food supplement production [54, 53]

Bioethanol derived from lignocellulosic sources like banana pseudostem plays diverse roles in the food industry. As a renewable and eco-friendly solvent, it is used to extract flavors, antioxidants, and bioactives for beverages, bakery items, and nutraceuticals. Its strong antimicrobial properties make it effective in sanitizing fruits, vegetables, meat, dairy,

and beverage processing environments, thereby enhancing food safety. Furthermore, bioethanol is incorporated into edible coatings and active packaging films to prolong shelf life by limiting microbial growth and moisture loss. It also functions as a green processing aid in the formulation of food additives, enabling efficient production without harmful chemical residues. These multifunctional roles position bioethanol as a cornerstone of sustainable, safe, and innovative food processing and preservation.

9. Comparative Yield Studies

Several studies have demonstrated significant improvements in ethanol yield using ultrasound-assisted or enhanced methods in banana biomass processing:

- a. Ingale *et al.* (2014) reported an ethanol yield of 17.1 g/L from banana pseudostem (BPS) through co-culture fermentation using *Aspergillus ellipticus* and *Aspergillus fumigatus* for saccharification, followed by *Saccharomyces cerevisiae* for fermentation. The synergy between fungal enzymes and microbial fermentation significantly boosted ethanol production [31]
- b. Kusmiyati *et al.* (2018) achieved 8.51 g/L ethanol using simultaneous saccharification and fermentation (SSF) of banana stem with a microbial consortium. This method utilized the inherent fermentable sugars and efficient biomass conversion, demonstrating the viability of banana pseudostem as a feedstock ^[15].
- c. Matharasi *et al.* (2018) attained the highest reported yield of 23.42 g/L ethanol from spoiled banana, optimized by adjusting inoculum concentration and fermentation pH. The study highlighted how controlled parameters could maximize ethanol output, particularly when using waste fruit biomass ^[55].

10. Challenges

Ultrasound-assisted, non-enzymatic pretreatment of banana pseudostem (BPS) offers considerable potential for sustainable bioethanol production; however, several obstacles continue to impede its large-scale industrial application. A key challenge lies in the high capital and operational costs associated with ultrasound technology, particularly due to the intensive energy demands of focused sonication systems. Moreover, variability in biomass characteristics-driven by regional, seasonal, and varietal factors-hampers process uniformity and scalability. Although chemical pretreatments can effectively increase cellulose accessibility, they often lead to the formation of fermentation inhibitors such as hydroxymethylfurfural, which reduce microbial activity and ethanol output [56].

Despite rising academic interest in ultrasound-based methods, there remains a significant gap in technoeconomic analyses and life cycle assessments (LCA), limiting our understanding of their commercial viability and environmental performance [57, 58].

To address these limitations, future research should prioritize the development of green, energy-efficient pretreatment strategies-including biological or hybrid approaches that combine ultrasound with enzymatic or microbial delignification. Additionally, genetic modification of microbial strains, particularly those capable of fermenting

both hexose and pentose sugars, could enhance substrate utilization and increase ethanol productivity [25].

Conducting comprehensive LCA and economic modeling will be critical in verifying the sustainability and cost-effectiveness of BPS-derived ethanol. Furthermore, integrating ethanol production within biorefinery systems-where value-added co-products like bioplastics, organic acids, and nutraceuticals are simultaneously produced-could significantly improve the economic feasibility of the process and support circular economy principles [59].

11. Future Scope

Despite encouraging laboratory-scale outcomes bioethanol production from banana pseudostem (BPS), several hurdles continue to obstruct its commercial-scale deployment. For instance, Legodi et al. (2021) reported ethanol yields of up to 17.6 g/L under controlled lab settings. However, scaling such processes to industrial levels is hampered by high pretreatment costs, feedstock inconsistency, and operational complexities [60]. Moreover, conventional acid and alkali pretreatment techniques are not only energy-intensive but also prone to generating fermentation inhibitors like furfural and HMF, which hinder microbial activity and reduce ethanol yields [56]. Another major bottleneck is the inefficient utilization of hemicellulosic pentose sugars, as traditional yeasts such as Saccharomyces cerevisiae preferentially ferment hexoses. Enhancing pentose conversion through engineering or co-culturing with pentose-fermenting microbes could substantially improve ethanol output [59]. Additionally, the lack of detailed techno-economic evaluations and life cycle assessments (LCA) limits the understanding of commercial feasibility and environmental implications. Rahman et al. (2019) emphasized the importance of comprehensive economic and environmental modeling to support industry-scale adoption [57].

Future research should prioritize the development of nextgeneration pretreatment technologies-especially biological or combined physicochemical methods-that can efficiently release fermentable sugars while minimizing the formation of inhibitory compounds [56]. Furthermore, engineering robust microbial strains or employing co-culture systems, such as Candida guilliermondii and Candida tropicalis, can enhance the co-fermentation of hexose and pentose sugars, thereby boosting ethanol yields [57]. Incorporating BPSbased bioethanol production within an integrated biorefinery framework also opens the door to co-producing value-added compounds like organic acids and bioplastics, improving both environmental and economic sustainability [59]. Lastly, full life cycle assessments are critical for quantifying the environmental footprint-such as greenhouse gas emissions and energy consumption-ensuring that bioethanol derived from BPS can serve as a genuinely sustainable and viable alternative to fossil fuels [60].

12. Conclusion

This review underscores the considerable potential of banana pseudostem (BPS) as a sustainable lignocellulosic resource for second-generation bioethanol production. Among the various pretreatment approaches examined, ultrasound-assisted, non-enzymatic methods-especially when coupled with chemical agents like NaOH and H₂SO₄-

emerge as particularly effective. These techniques significantly enhance cellulose accessibility and the release of fermentable sugars. The application of ultrasound facilitates structural disruption of the lignocellulosic matrix, increases fiber porosity and crystallinity, and improves thermal properties, resulting in improved ethanol yields without the high costs associated with enzymatic hydrolysis. When paired with microbial co-culture systems or genetically engineered fermentation strains. pretreatments offer a cost-effective and scalable strategy for converting agricultural waste into renewable biofuels. As such, ultrasound-assisted processing holds strong promise for supporting circular bioeconomy objectives while also addressing challenges related to agro-waste utilization.

Nevertheless, several key challenges must be addressed for industrial-scale adoption. Energy consumption associated with ultrasound systems-particularly high-intensity focused sonication-can be substantial, and if not precisely controlled, may lead to cellulose degradation. Chemical pretreatments, although efficient, can generate inhibitory by-products that compromise fermentation efficiency. Additionally, the limited ability of Saccharomyces cerevisiae to ferment pentose sugars remains a constraint, despite advances in metabolic engineering and co-culture strategies. Economic assessments of large-scale production remain insufficient, with a lack of comprehensive data on life cycle impacts, cost-effectiveness, and process energy demands. Moreover, integration of BPS valorization into broader biorefinery frameworks-such as those enabling co-production of bioplastics, organic acids, or biogas-is still in early stages. To fully realize the industrial potential of ultrasoundassisted BPS bioethanol production, future efforts should focus on optimizing pretreatment parameters, enhancing microbial fermentation efficiency, conducting in-depth environmental and economic assessments, and developing integrated biorefinery models. Such advancements are critical to transforming this promising laboratory-scale process into a viable, sustainable, and commercially competitive biofuel technology.

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