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Ecologically Clean Fuel from Common Cane

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ABSTRACT: Second-generation bioethanol produced from lignocellulosic materials such as wood, agricultural or forestry residues has the potential to replace or replace expensive gasoline. One of the most important steps in ethanol production is the hydrolysis of hemicellulose and cellulose into monomeric sugars. The most promising method of hydrolysis of cellulose to glucose is the use of enzymes. However, some pre-treatment is required to make the raw material enzyme-friendly. The whole process must be taken into account when evaluating the processing efficiency, since different pretreatment methods allow the production of different types of materials. This affects how the enzymatic hydrolysis should be carried out how the lignin is extracted and the use of the lignin by-product. Biofuels produced from lignocellulosic materials, called second-generation bioethanol, have energy, economic and environmental advantages over starch or sugar bioethanol.

KEY WORDS: bioethanol, pretreatment, common cane, lignocellulosic feedstock, cellulose, glucose.

I. INTRODUCTION

Bioethanol is a biodegradable, high-octane motor fuel made from cellulose found in plants. Bioethanol production using second-generation plant lignocellulosic material biomass as a substrate is promising for an environmentally friendly environment. The most common, cheap and renewable energy source for bioethanol production is plant biomass lignocellulosic material. For the production of bioethanol, lignocellulose obtained from second-generation biomass requires pre-treatment by physico-chemical, mechanical, chemical and biological methods. Pretreatment reduces the complex nature of the structures to simple monomers and then converts them into bioethanol using ethanol-producing microorganisms. Pre-treatment is mainly required to increase cellulose permeability by increasing the availability of enzymes. The complex nature and ultrastructure of second-generation lignocellulosic biomass is due to the linear glucose molecule tightly bound to the lignin molecule, which prevents the presence of the glucose molecule. Pretreatment removes the lignin molecule, making this glucose molecule available for ethanol production. In order to maximize the positive benefits of biofuels and minimize their negative effects, a consistent scientific basis must be properly sequenced. Lignocellulosic materials are of different types depending on the pretreatment process and the conditions chosen for hydrolysis [1].

Currently, biofuel is attracting great interest worldwide due to its environmental friendliness. Biofuel is a renewable, carbon-neutral source that does not disrupt the air balance in the atmosphere, which causes global warming. Biofuel is one of the most possible ways to free people from dependence on traditional fossil resources [2].

Over the past 20 years, the crude oil crisis has increased the demand for renewable energy, especially biofuels. America and Brazil are the two leading countries in the production of bioethanol from sugarcane and corn. According to a 2009 report, Brazil produced 12.5 billion liters of bioethanol annually from sugarcane as an industrial fuel, while in America 5 billion liters of bioethanol were produced from corn, 111 gas stations sold E85 gasoline (gasoline blend 85 % vol. consists of bioethanol) [3]. However, the production of biofuels from starches and sugars seriously undermines global food security for humanity.

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Table 1.
Comparison of starch with cellulose and glycogen

Naming	Starch	Cellulose	Glycogen
Existence in nature	Widely distributed in plants as a carbohydrate store	The main component of the plant cell wall	Found in fungi and animals as energy sources
A monomer unit	Alpha-glucose	Beta-glucose	Alpha-glucose
Chemical structure of the polymer	1,4-glycosidic bonds in amylose; 1,4- and 1,6-glycosidic bonds in amylopectin	1,4-glycosidic bonds	1,4 and 1,6-glycosidic bonds
Molecular morphology	Amylose in the form of an unbranched helical chain; Amylopectin is in the form of a long branched partially helical chain	A straight, long, unbranched chain with H-bonds between adjacent chains	Short, multi-stranded chains, partially coiled
Average molecular mass	Changeable	162.14 g/mol	666.6 g/mol

This simple process involves two main steps. First, lignocellulose must be pretreated to remove lignin and enhance the penetration of hydrolysis agents without chemical degradation of cellulose and hemicellulose. Second, the pretreated material is converted into bioethanol through hydrolysis and fermentation. Also, some unique published studies and promising processing methods for biomass to bioethanol conversion and economic efficiency are briefly described.
However, physical and chemical barriers resulting from the close association of the main components of lignocellulosic biomass prevent the hydrolysis of cellulose and hemicellulose into fermentable sugars. The main purpose of pre-treatment is to increase the availability of enzymes that improve the pulping. Each pretreatment has a specific effect on the cellulose, hemicellulose, and lignin fractions, so different pretreatment methods and conditions must be chosen according to the process configuration chosen for the subsequent hydrolysis and fermentation steps.

II. SIGNIFICANCE OF THE SYSTEM

This article reviews the most interesting technologies for ethanol production from lignocellulosic, exploring several key properties that should be targeted in low-cost and advanced pretreatment processes. The Methodology and Discussion is presented in section III, section IV covers the experimental results of the study, and section V discusses the future study and conclusion.

III. METHODOLOGY AND DISCUSSION

Inexpensive sources of lignocellulosic biomass can be waste from forestry, agriculture and agro-industrial complex. Many such materials can be mentioned here, including sawdust, cottonwood, sugarcane stalks, beer dregs, grasses and straws, grain, corn, and barley stalks, leaves, husks, core and skins. Contrary to the desire to use these materials for the production of valuable products, lignocellulosic waste accumulates in large quantities every year, causing environmental problems [3].
Lignocellulose consists of cellulose, hemicellulose and lignin and is always present together with other extracts and mineral residues. The total composition of lignocellulose is presented in Table 2. In lignocellulose, strands of cellulose fibers are formed by linking cellulose together using hydrogen bonds. The structure of cellulose in the polymer is not homogeneous. The crystalline area is where the cellulose nanofibers are organized and dense, while the amorphous area is irregular and easily hydrolyzed [4]. Cellulose fibers are skeletons surrounded by hemicellulose and lignin (Figure 1). This structure naturally protects polysaccharides from hydrolysis by enzymes and chemicals, which makes it difficult for the chemical and bioconversion of lignocellulose to other products, namely ethanol.

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Table 2.
Specific chemical composition of lignocellulosic biomass

Raw material	Lignin (мas.%)	Cellulose (мas.%)	Hemicellulose (мas.%)
Leaf type	18–25	45–55	24–40
Conifers	25–35	45–50	25–35
Drugs	10–30	25–40	25–50

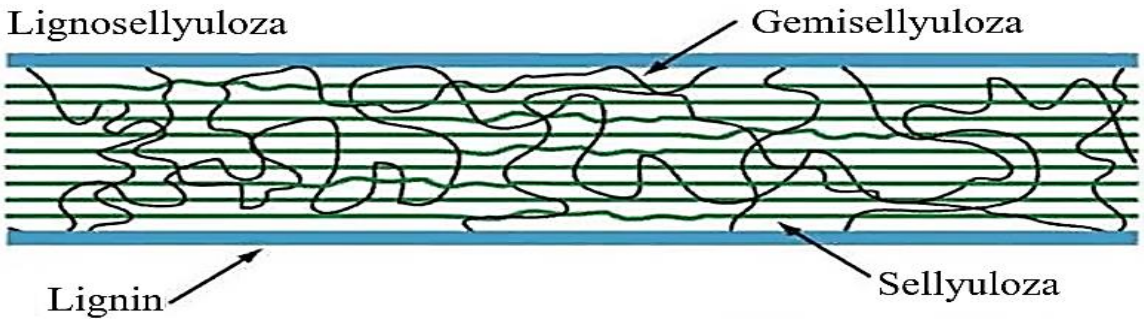

Fig. 1. Material structure.

In addition to cellulose, hemicellulose is a well-known polysaccharide in lignocellulose. Hemicellulose is a linear and branched heterogeneous polymer, usually composed of five different sugars—L-arabinose, D-galactose, D-glucose, D-mannose, and D-xylose. The basis of hemicellulose chains can be homopolymer or heteropolymer (a mixture of different sugars). Hemicelluloses differ from cellulose not only by different sugar units, but also by their amorphous molecular morphology, where short chains branch off from the main chain molecules. Due to this chemical classification, hemicellulose is more easily hydrolyzed than cellulose [5].
Lignins play the role of cross-linking cellulose and hemicellulose in the matrix. Lignins can be dissolved in acidic and alkaline solutions, their solubility depends on their previous state. These properties of lignins make lignocellulose structure stronger and tougher in nature, resisting hydrolyzing agents. Therefore, pretreatment to remove lignins from lignocellulose and enhance the penetration of hydrolysis agents is an important step in the process of converting biomass to bioethanol [6].
For the general chemical expression of the conversion of natural glucose-based carbohydrates to bioethanol, it can be expressed as follows:

$$(C_6H_{10}O_5)n + nH_2O \rightarrow nC_6H_{12}O_6 \rightarrow 2nC_2H_5OH + 2nCO_2$$

Pretreatment processes are applied before the hydrolysis and fermentation of lignocellulose. In this:
- It is necessary to increase amorphous spheres, which are easier to hydrolyze than crystalline cellulose;
- Porosity of fibrous matrices should be increased to ensure the penetration of chemicals and enzymes into the structure;
- Cellulose is removed from the medium of lignins and hemicelluloses.

Physical, chemical and biological pretreatment methods are used in pretreatment of lignocellulose. In order to classify the physico-chemical methods of preliminary processing, they are briefly described below:

- **Mechanical processes** reduce the volume of biomass and thereby increase the contact surface. Mechanical processes do not change the chemical properties of materials. Therefore, they can be a raw material processing step before other pre-processing steps. Cutting, grinding, milling and grinding can be done using special equipment.

- **Thermophysical methods:** grinding, steam explosion, high pressure steaming, etc. can improve the contact surface, reduce the degree of polymerization of cellulose, reduce the crystallinity of cellulose, and cause the cross-linking of lignin to crack slightly.

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- **Critical CO_2 Extraction Method:** At first glance, this method was expected to produce a pulp pack in the lignocellulosic pretreatment. However, the cost of the system is too high for high-pressure equipment, making it unsuitable for industrial production. In addition, other similar studies have shown that this method is not effective in increasing the bioethanol yield of biomass [7].

- **Ionic liquid methods:** ionic liquids are also known as liquefied organic salts, which have several special properties such as volatility, high thermal stability, and especially cellulose solubility. Therefore, ionic liquid has been studied for pretreatment of lignocellulose as a green solvent.

Pretreatment with organic solvents and surfactants for lignin removal is of some interest, but the results are not only for academic research or only for lignin extraction [8].

Chemical methods

The use of aqueous acidic and alkaline solutions depends on the solubility of lignin in them. Lignocellulosic pretreatment with acid and alkali is still the most effective and most acceptable technology for lignocellulosic bioethanol production technology [9]

Acid pretreatment: H_2SO_4 and HCl are widely used for lignocellulose pretreatment. Due to the toxicity and hydrolysis of cellulose and hemicellulose during pretreatment, highly concentrated acidic solutions are not preferred. In addition, when using highly concentrated acids, it has a corresponding effect on the corrosion of the equipment and the safety of human operators. Diluted acids are also not recommended due to the formation of furfural compounds during the initial treatment, which inhibit the growth of microorganisms during fermentation.

Alkaline pretreatment: Compared with acids, pretreatment of lignocellulose with alkaline solutions minimizes the loss of carbohydrates due to hydrolysis. Also, alkali helps remove acetyl groups, promotes further hydrolysis and inhibits furfural formation [10]. Sodium hydroxide is the most suitable alkali solution due to its low cost and high efficiency in pretreatment of lignocellulose.

Combination of pre-processing methods: It can be seen that a single pre-processing method cannot produce the expected result. Therefore, the combination of pretreatment methods has become a common procedure in the production of bioethanol from lignocellulosic. First, lignocellulose must be reduced in size by mechanical processes such as milling, cutting, and shredding of the biomass, optionally steam explosion before soaking in an alkaline or acidic solution (alkaline pretreatment is preferred here). Second, the pre-treated material is washed and neutralized. The final product of this stage is a hardened material that is always ready for further processing [11].

IV. EXPERIMENTAL RESULTS

Pretreated biomass can be directly converted to bioethanol by microbial conversion (CMC) or by hydrolysis with fermentation. In fact, the TMC method is time-consuming, and the conversion efficiency is very low when the risk of contamination is high. In contrast, microbial fermentation, generalized enzymatic hydrolysis, is the preferred method, which has been shown to perform much better [12].

Saccharification of lignocellulose. After initial processing of lignocellulose, the polysaccharide-enriched material is hydrolyzed by enzymes into single sugars (hexose and pentose). Commercialized enzyme for the hydrolysis of cellulose and hemicellulose is actually a mixture of different types of enzymes extracted from a microorganism, usually called cellulose. These enzymes usually invert or cleave glycosidic bonds in carbohydrates through retention mechanisms, the latter occurring via a two-step mechanism involving the formation of a glycosyl-enzyme intermediate [13].

Unlike hemicellulose, which is more easily hydrolyzed, cellulose has a stable crystal structure that resists depolymerization. For effective enzymatic hydrolysis of cellulose, the following three cellulose enzymes are widely used [14].

- Endo-1,4-β-glucanases (EG) or 1,4-β-D-glucan 4-glucanohydrolases (EC3.2.1.4). This enzyme randomly breaks 1,4-β-glucan bonds.

- Exo-1,4-β-D-glucanases, including 1,4-β-D-glucan-gluco-hydrolase (EC 3.2.1.74), cleave D-glucose from 1,4-β-D-glucan and D -slowly hydrolyzes cellobiose, the enzyme 1,4-β-D-glucan cellobiohydrolase (EC 3.2.1.91) (CBH) cleaves cellobiose from 1,4-β-glucan.

- β-D-glucosidase or β-D-glucoside glucohydrolase (EC 3.2.1.21) to produce D-glucose from cellobiose and other glucose oligomers.

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Enzymatic hydrolysis favors the growth of microorganisms during fermentation, although there is little difference in the optimum temperature for the two processes.

Fermentation. Microorganisms are used to metabolize some of the released sugars into bioethanol through enzymatic hydrolysis. There are two approaches:

- **Separate Hydrolysis and Fermentation (Sh and F):** Hydrolysis is carried out to the end, then microorganisms are added to the mixture to ferment the sugars. This method has its own weaknesses, including contamination, the formation of inhibitors, and the need for more time and additional equipment;
- **Simultaneous saccharification and fermentation**: enzymatic hydrolysis and microorganism fermentation are carried out simultaneously in one equipment. Both enzymes and microorganisms are loaded into the mixture. This method has been proven to be more efficient than the AGF described above, with shorter time, less equipment, and minimal risk of contamination.

Currently, simultaneous saccharification and fermentation is the optimal way to convert lignocellulose into bioethanol. This process is characterized by high conversion efficiency. However, this method still has minor drawbacks. The optimal temperature for enzymatic hydrolysis is 45-50°C, the highest efficiency of fermentation is 28-35°C. In addition, some intermediates also resist the growth of microorganisms [15].

Various microorganisms can be used to enhance fermentation. Table 3 lists some of the most popular microorganisms that metabolize sugars and are used in ethanol production.

Table 3.
Some popular microorganisms for bioethanol production

Description	Microorganism			
	E. coli	Z. mobilis	Saccharomyces cerevisiae	Pichia stipitis
D-glucose fermentation	+	+	+	+
Other uses of hexose (D-galactose and D-mannose)	+	–	+	+
Recovery of pentoses (D-xylose and L-arabinose).	+	–	–	+
Direct use of hemicellulose	–	–	–	w
Anaerobic digestion	–	+	+	w
Formation of mixed product	+	w	w	w
High ethanol yield (from glucose)	–	+	+	w
Ethanol tolerance	w	w	+	w
Tolerance to lignocellulosic inhibitors	w	w	+	w
Acidic pH range	–	–	+	w

[+]positive,[-]negative, [w] weak

From the data of Table 3, it is easy to see why Saccharomyces cerevisiae is a suitable yeast choice for converting sugar solution into bioethanol. Saccharomyces cerevisiae is not only a well-known traditional yeast, but also the most popular yeast in bioethanol production due to its tolerance to high ethanol concentrations and material inhibitors.
During the fermentation process, additional nutrients can be added to provide an organic nitrogen source for the growth of microorganisms.

V. CONCLUSION AND FUTURE WORK

Lignocellulosic bioethanol production can meet the requirements of food security and the sustainable vision of a green world. The process includes pre-treatment, enzymatic hydrolysis and fermentation steps. All over the world, intensive

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research is being done to increase the economic efficiency of ethanol production and to transfer it from laboratory to industrial/commercial scale.

The production process of cellulosic bioethanol includes certain processing steps, especially pretreatment and hydrolysis. Microorganisms adapted to ferment C_5 and C_6 sugars are needed and will need to be studied.

New combined processes reduce both the number of work steps and the production of chemical inhibitors. Recent advances in the field of genetically modified Saccharomyces cerevisiae offer many opportunities to improve alcohol tolerance and conversion efficiency. If existing processing problems are overcome and the best combination of advanced systems is used, second-generation bioethanol can outperform conventional first-generation processes.

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