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Qualitative Study of the Pre-Treatment Stage in Ethanol Production from Lignocellulosic Biomass

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ABSTRACT: *Global warming, energy dependence on fossil fuels and exponential demand for space for food production has raised interest in alternative renewable energy sources, among them the second generation of bioethanol better known as lignocellulosic bioethanol, a biofuel from biomass. The research performed in this article consists of a bibliographic review about bioethanol. The general objective of this research is to present a qualitative study about the different stages of pre-treatment of biomass in the production of lignocellulosic bioethanol, as well as to know the different routes of its obtaining. With this study, it was possible to enumerate and compare the advantages and disadvantages of pre-treatment processes already implemented. Thus, it could be concluded that this sustainable alternative is beneficial, but lack incentive in future research.*

KEYWORDS *Ethanol, lignocellulose, pre-treatment.*

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I. INTRODUCTION

The incentive and ease of having a light vehicle led to a significant increase in the vehicle fleet and consequently growth in fossil fuels consumption. However, to meet demand, Brazil needs to import gasoline and diesel, as the country's oil refining capacity is limited. Therefore, it is speculated that this import will reach even larger volumes over the years (MILANEZ et al., 2015).

Furthermore, there is an economic concern about the instability of the price of a barrel of oil, which is dependent on several factors for its appreciation. These include: physical and chemical characteristics, value of raw materials, political incentive in production, occurrence of conflicts in production areas and fuel performance. Each country has its regulator and each fuel has its parameter specification, where regulators have limited freedom in setting parameters and control ranges.

However, due to the acceleration of the concentration of carbon dioxide (CO_2) in the atmosphere, the limitation of oil and the constant instability in the value of oil, there was a need to consolidate an independence of its products. For this reason, the development and implementation of the use of renewable energies such as biofuels, wich replace fossil derivatives, is increasingly being sought.

Biofuels are derived from renewable biomass, capable of partially or wholly supplying petroleum and natural gas fuels in combustion engines or other power generation. In the market, there are two biofuels with greater consolidation and acceptance: the biodiesel, which competes with petrodiesel, and bioethanol, a competitor of gasoline, and sugarcane fermentation is its main source of production in Brazil.

Brazil is at the forefront of the world in terms of relative consumption, being the country with the largest relative substitution of gasoline for ethanol in the world, where E5 gasoline has been standardized since 1931 (MAPA, 2015). Currently, retail outlets have, for consumer’s choice, two types of Otto cycle fuels: hydrous ethanol – HE and gasoline E27 (gasoline blend and anhydrous ethanol – AE) (LOPES NETO et al., 2019). Table I, with data from the Brazilian Statistical Yearbool of Oil, Natural Gas and Biofuels 2019 (ANP, 2019) shows the evolution of Otto cycle fuel consumption in Brazil.

Table I – Evolution of Otto cycle fuel consumption in Brasil 2009 – 2018.

Combustível	**2009**	**2010**	**2011**	**2012**	**2013**	**2014**	**2015**	**2016**	**2017**	**2018**
Gas (10^6 m^3)	19,06	22,76	27,07	31,76	31,68	33,27	30,20	31,40	32,23	28,00
AE (10^6 m^3)	6,35	7,09	8,20	7,94	9,69	11,09	10,93	11,62	11,62	10,35
Blend Gas–AE	E25	E20 E25	E25 E20	E20	E20 E25	E25	E25 E27	E27	E27	E27
HE (10^6 m^3)	16,47	15,07	10,90	9,85	11,75	12,99	17,86	14,59	13,64	19,38
Ethanol (AE +HE) (10^6 m^3)	22,82	22,16	19,10	17,79	21,44	24,09	28,80	26,20	25,26	29,74
Ratio ethanol/gas	1,198	0,974	0,706	0,560	0,677	0,724	0,953	0,834	0,784	1,062

Gas: Gasoline; AE: anhydrous ethanol; HE: hydrous ethanol.

As can be seen in Table I, over the period surveyed there was an increase in gasoline consumption, which is linked to the increase in the Brazilian vehicle fleet; the increase in anhydrous ethanol is connected with the vehicle fleet and the gasoline-ethanol blend ratio. However, the proportion of the gasoline-ethanol blend is dependent on ethanol supply in Brazil, linked to complex aspects such as: sugarcane crop size, distribution logistics, climatological aspects, productivity of sugarcane plants, international price of sugar etc. In turn, the consumption of hydrous ethanol is linked to its supply and the prices of this biofuel and gasoline. For example, between 2011 and 2014, Brazil, through Petrobras, maintained a policy of controlling low gasoline prices, causing little competitiveness of hydrous ethanol. From 2015, there was a change in this policy and an increase in the crop, causing increases in the EA content in the gasoline-ethanol blend and in the consumption of HE. In 2018 for example, total ethanol consumption (AE+HE) exceeded gasoline: $29{,}7 \times 10^6$ m^3 and $28{,}0 \times 10^6$ m^3, respectively.

Therefore, to increase total ethanol consumption, increasing the sustainability of the Otto cycle vehicle fuel matrix, it is necessary to increase the supply and the productivity of the total ethanol production processes, in order to guarantee a continuous flow of bioethanol and competitive prices.

Today, others are reevaluating their energy policies, making them more sustainable. A survey that identifies biofuel and biofuel blending mandates around the globe suggests different motivations for adopting these blends, ranging from concerns about reducing greenhouse gas emissions – GHG to the energy security of countries. Fig. 1 shows the percentage of ethanol addition that some countries adopt.

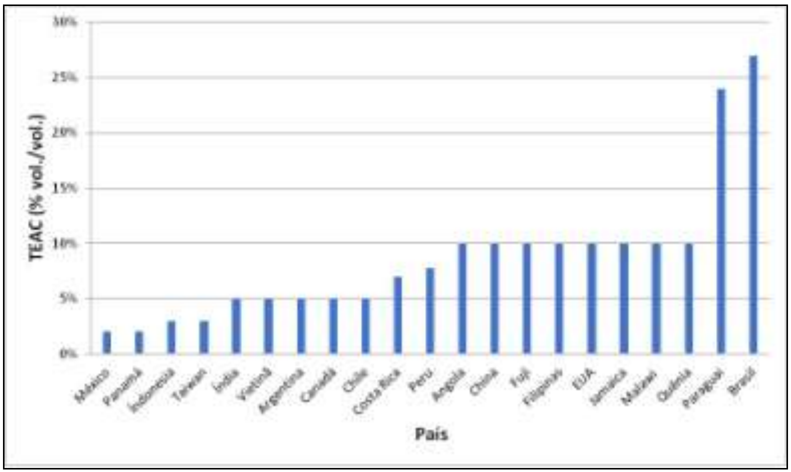

Fig. 1. Gasoline/ ethanol blends in some countries.

Source: Adapted from Lane (2019).

The use of gasoline EXX blends is observed in several countries. For bioethanol percentage growth it is necessary to increase production, so the lignocellulosic pathway is a new and promising route.

II. BIOETHANOL OBTAINED BY FERMENTATION

Ethanol is colorless, highly flammable compound of the formula C2H5OH, found in the liquid form in the STP (Melting Point at -173,38 ºF and Boiling Point at 172, 94 ºF), (which may ignite if subjected to a spark from 55,4 ºF). In addition, it has a characteristic odor, is volatile and water soluble. It is toxic in its pure state and in aqueous solutions with low concentration can be ingested by humans in a moderate way (PACHECO, 2010; ANP, 2016; NOVA CANA, 2016).

Ethanol can be obtained by petrochemical route (ethylene hydration), whose process is little used in Brazil. However, it is estimated that 80% of ethanol produced in the United States is from ethylene hydration (NOVA CANA, 2016).

When ethanol is obtained from biochemical processes, it is called bioethanol, with three distinct classes of procedures, called by the researchers as “bioethanol generations”. The main way to obtain bioethanol, called first generation (ethanol 1G or E1G), is from the fermentation of sugar-rich biomass (sucrose or starch), such as corn, sugar cane, sugar beet, wheat straw rice etc.

Second generation ethanol (E2G) is currently in its initial production phase, the main object of this study, also known as cellulosic ethanol or lignocellulosic ethanol, which is derived from plant biomass. ethanol 3G (E3G), still in its initial phase of studies, uses marine biomass, macro-algae or micro-algae, as a source of raw material.

The first generation of bioethanol is the most widespread and known form of biochemical ethanol production, occurring from the conversion of sugars or starch. The production process consists of a sucrose hydrolysis step, followed by alcoholic fermentation of glucose.

Alcoholic fermentation is an anaerobic reaction, catalyzed by microorganisms that degrade sucrose into glucose and fructose; They then convert one glucose molecule into two ethanol molecules and two carbon dioxide molecules. The microorganisms used for this conversion of sugar into ethanol are called yeast, which constitute a group of unicellular beings of asexual reproduction and capable to fermenting sugars present in aqueous solution in the absolute absence of oxygen (ZÚÑIGA, 2010).

An example of yeast used in fermentation is Saccharomyces Cerevisiae. It is a special yeast for alcoholic fermentation, that is, it is an optional anaerobic responsible for the production of enzymes fundamental for the breakdown of glycoside molecules in monosaccharides by the invertase enzyme. Subsequently, these monosaccharides are subjected to the action of another enzyme, called zymase which in turn produces ethanol (PACHECO, 2010; MENDES; DILARRI; PELEGRINI, 2015).

In Brazil, sugarcane is the main raw material in the production of first generation ethanol and its residue, bagasse, can be used to produce energy or ethanol 2G. sugar cane is used because it has high carbohydrates concentration, low relative lignin content, easy use, low cost of harvesting, transportation and storage (GOUVEIA et al., 2009).

Bioethanol obtained by fermentation of sugar cane

In Brazil, bioethanol production plants from sugar cane are know as mills and a significant part is integrated into unit operations for sugar production, as can be seen in Fig. 2 below.

Each ton of sugar cane can produce up to 80 liters of bioethanol (NOGUEIRA, 2004). The agroindustrial process involves the harvesting of sugar cane, mechanized in several farms, transportation to mills and the washing process to remove impurities. Then the milling step is directed, where the stem of the plant is crushed by rolls in order to obtain the juice, known as molasses, which corresponds to 70% (m/m) of sugar cane. Soon after, the molasses is sent to a settling tank, to eliminate the remaining impurities; then the purified broth is sterilized. (NOVA CANA, 2019).

purified broth may be used to produce sugar or ethanol; thus, sugarcane industry plants are designed to produce ethanol or sugar. residual bagasse can be used as furnace fuel for heat or energy production, or for 2G ethanol production.

In Brazilian mills, the sterilized broth is directed to the fermentation tanks, where it can receive water and molasses to obtain a total concentration of fermentable sugars of 180 g L-1 of sucrose. molasses is a byproduct of sugar production, which contains carbohydrates (around 45 – 60% sucrose and 5 – 20% glucose and fructose), which can be used to enrich the sugar content of the broth in the fermentation tank (BASSO et al., 2008).

Still in the fermentation tank, the addition of Saccharomyces Cerevisiae yeasts causes fermentation, where the microorganisms catalyze the biochemical reaction, lasting an average of 6 to 12 hours. At the end of the period, the broth is transformed into fermented wine, with ethanol content in the range of 7% to 10% v/v. It should be mentioned that higher concentrations of ethanol, poison the yeast catalysis, paralyzing the reaction (DUARTE; LOURENÇO; RIVEIRO, 2006; NOVA CANA, 2019).

Subsequently, the mixture is conducted to distillation columns, in order to separate the ethanol. After two successive distillations, hydrated ethanol, azeotrope ethanol/water, with an alcohol content of 96% m/m (93% v/v) is obtained. If you wish to increase the purification, hydrous ethanol is directed to dehydration columns, where anhydrous ethanol is obtained, with an alcohol content of 99.5% m/m ((NOVA CANA, 2019).

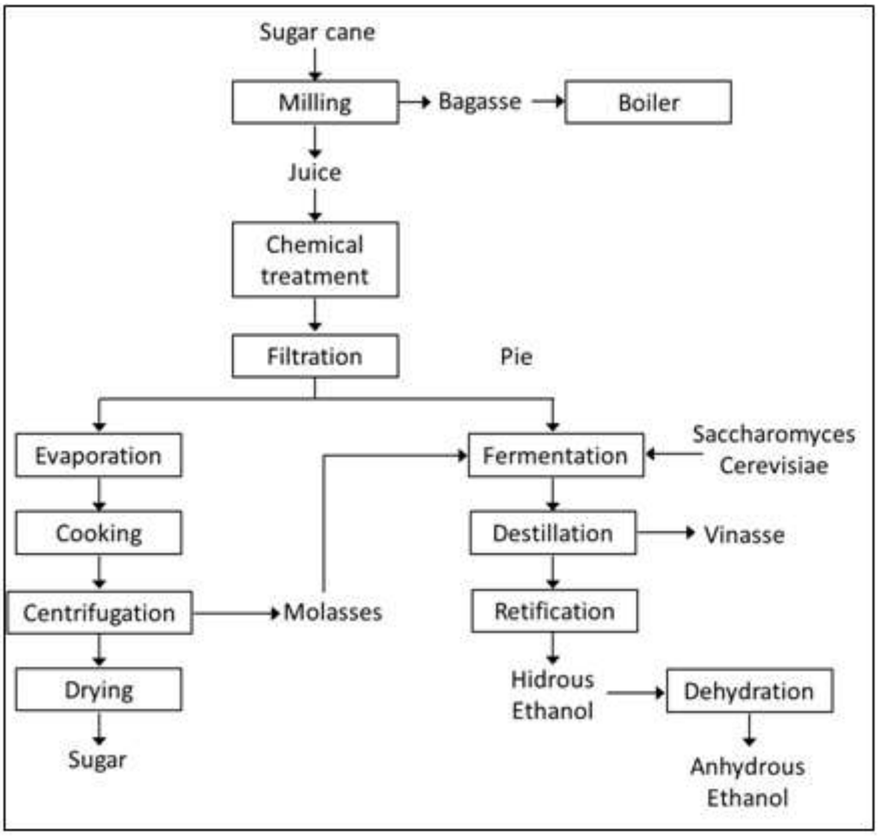

Fig.2. Flowchart of sugar and ethanol production.
Source: Adapted from Cruz (2019).

When alcoholic fermentation is conducted at 93,2 °F, ethanol concentrations from 40.0 g L-1 decrease yeast enzymatic activity; concentrations close to 95.0 g L-1, inhibit yeast growth. At higher temperatures, due to the increased d cell membrane fluidity, ethanol has a higher yeast toxicity effect. At lower temperatures, yeast resistance to ethanol is increased and yeast catalytic activity can be obtained at ethanol concentrations higher than 95.0 g L-1 (VELOSO; RODRIGUES; BADINO JÚNIOR, 2018). Thus, processes conducted with low temperature controls can generate wines with higher alcohol content, optimizing the productivity and ethanol recovery costs in distillation.

Two other fields of research to optimize bioethanol fermentation productivity deserve to be highlighted: the prospecting of robust yeast strains, with high tolerance to ethanol and temperature, capable of acting in more aggressive media, in order to produce fermented wines of high alcohol concentration (MIRANDA JÚNIOR et al., 2009); fermentation technology in systems with a high concentration of fermentable sugars (VHG), which has additional advantages of saving water, reducing costs with vinasse treatment, saving energy on distillation and reducing bacterial contamination (QIU; JIANG, 2017).

Therefore, recent research, combining two more different parameters, indicates the existence of fertile areas for increasing the productivity of alcoholic fermentation. Cruz (2019) evaluated the performance of different Saccharomyces cerevisiae strains at different temperatures (68 ºF to 86 ºF), under VHG conditions and in repeated fermentation batches; the best system provided ethanol concentration productivity values of 146.7 g L-1 employing 80,6°F temperature and 300 g L-1 sugars concentration in the fermentation tank.

Bioethanol obtained from corn fermentation

The production of bioethanol from corn begins by cleaning the post-harvest grains by separating the ear and stem. Subsequently, the grains are directed to the, mechanical and thermal disintegration process, where they are ground and cooked, in boiling or steaming water to gelatinize the starch for enzymatic saccharification. This will reduce sugar chains 15% (ALCARDE; BORTOLETTO, 2015).

Then, the saccharified wort is deposited in the fermentation tank, where it receives the addition of yeast for the fermentation process. In older processes, fermentation time is longerraging from 54 to 72 hours (LIMA et al., 2001); modern corn fermentation processes have an average duration of 28 to 30 hours (ALCARDE; BORTOLETTO, 2015). However, fermented wine, in both processes, has a higher concentration of ethanol, compared to traditional sugarcane processes, reaching 12 to 18% v/v (LIMA et al., 2001).

The mixture is distilled, resulting in two phases: crude ethanol and vinasse; ethanol is rectified (elimination of impurities) and dehydrated. final ethanol is stored for future use as fuel and vinasse (corn residue and dead yeast cells) is processed for animal feed (ALCARDE; BORTOLETTO, 2015). This is the main process of obtaining bioethanol in the USA.

One of the advantages of corn, as the raw material for ethanol production, is the ease of storage, since corn grains can be stored in silos for months, and consequently fermentations can be performed 345 days a year. In contrast, sugarcane needs to be milled immediately after harvesting to prevent microbial contamination, so that sugarcane mills can operate for 200 to 240 days annually (WHEALS et al., 1999).

This feature optimizes the distribution logistics of bioethanol, as it allows a better planning of ethanol production and dispenses tanks for storage during the post-harvest months, where there is no production in the plants.

Bioethanol obtained by fermentation of sugar beet

Sugar beet or white beet is used throughout Europe to produce ethanol. Before starting this process, the beets are removed from the ground and shaken to set aside. They are then washed and cut into sticks, to increase the contact surface and the extraction of sucrose in the hot water diffuser; later, the sticks are pressed until they reach 75% humidity. The pressed sticks are intended as animal feed and the liquid extracted from the pressing is destined for the solution contained in the diffuser and subsequently evaporated to concentrate the contained sugars. The concentrate (syrup) is directed to the alcoholic fermentation stage, in a process similar to that of sugar cane (BRASIL, 2008; BASSAM, 2010).

Despite the importance of the described processes, as mentioned above, this article focused on the second generation of bioethanol (E2G) production, as its growth estimate was considered in the coming years, both in the research area and in the implementation of new production plants on a commercial scale.

III. LIGNOCELLULOSIC BIOETHANOL – E2G (SECOND GENERATION)

Lignocellulosic bioethanol comes from the cell wall disruption of plant biomass, thus having access to cellulose so that it can be converted into fermentable sugars capable of generating bioethanol.

Cell walls are very complex structures that have a wide range of functions during all stages of plant life, such as providing mechanical robustness to cells; maintain its morphology; control both cell expansion and intercellular transport; in some cases it works as a food reserve; form protoplasts (plant cell components when devoid of cell wall); besides protecting the cell against most potentially pathogenic and predators organisms, which makes the production of lignocellulosic ethanol more complicated (TAIZ; ZEIGER, 2002).

The plant cell wall structure is subdivided into primary, secondary walls (being subdivided into S1, S2 and S3) and the middle lamella. These layers are predominantly composed of cellulose with a thickness of the order of 5 μm. The Cells are separated by the middle lamella, which is a thin layer, with a maximum thickness of 1μm, composed by a high concentration of lignin (GLÓRIA; GUERREIRO, 2006).

Lignin is, after cellulose and hemicellulose (low molecular weight polymerized monosaccharides with cellulose-like structure), one of the most abundant plant macromolecules found in the cell wall. Lignin is an amorphous, hydrophobic molecule tightly bound to hemicellulose and microfibrils, which makes lignocellulosic compounds resistant and recalcitrant to conversion by microbial and chemical agents (FUENTES, 2009).

However, there is concern about the manipulation of lignin biosynthesis pathways because it is an inhibitor in the fermentation step. This inhibition occurs due to the complexity of its molecule and the mechanical and chemical resistance to the use of cellulose and hemicellulose during bioethanol production. In an overview, Fig. 3 provides a simplified scheme of this process.

In the commercial route used in production plants, such as Granbio (an industry with an E2G plant called Bioflex 1 in operation since 2014, in the state of Alagoas-Brazil) which handles the straw or bagasse of traditional sugar cane or vertix-cane (biomass developed by genetic crossing of sugarcane ancestors that offers 3 times more productivity) as the main raw material (GRANBIO, 2019). According to Buckeridge, dos Santos and de Souza (2007), the process begins with the cell wall disruption through a stage called pre-treatment. In this stage, the objective is to remove structural and compositional barriers of lignocellulosic materials for use of polysaccharides as a source of fermentable sugars (hexoses and pentoses).

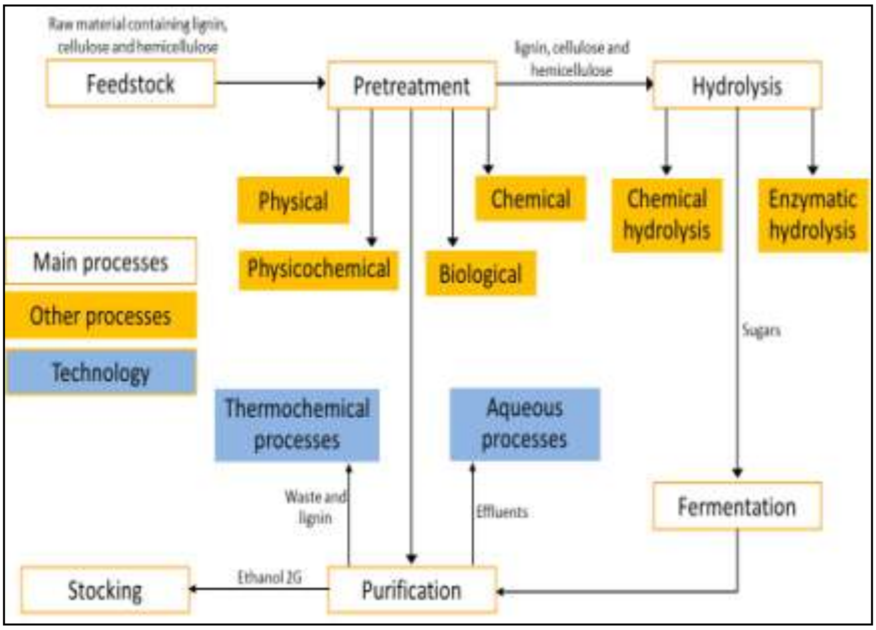

Fig. 3. General Process– ethanol production 2G.

Source: Adapted from Quilhó (2011).

However, due to this complex structure of the wall, the disassembly process must be as delicate as possible to preserve unharmed monosaccharides that will be used for fermentation. If the degradation is intense, furfural compounds are formed which are toxic to the yeast that will be used in the fermentation step. Thus, by hydrolyzing a mixture of cellulose and hemicelluloses, the temporal disconnection of glycosidic bond breakages of each type of polysaccharide becomes a barrier to the production of fermentable monosaccharides (BUCKERIDGE; DOS SANTOS; DE SOUZA, 2007). Subsequent steps involve the distilling the fuel for the purpose to removing any impurities still contained therein and the distributing the product as fuel ethanol.

In Brazil, sugar cane bagasse (BC) is the main raw material in the production of first and second generation ethanol. BC is used because it has a high carbohydrates concentration, low relative lignin content, easy use, low cost of harvesting, transport and storage (GOUVEIA et al., 2009).

IV. PRE-TREATMENT STEP

The pre-treatment step is the cell wall disruption without harming the molecule, so that a better efficiency occurs during hydrolysis. The use of different methods allows a better visualization of ethanol production in relation to enzymatic hydrolysis rates, fermentation load and variables as well as its economic viability and scale production (MANFREID et al., 2018).

Thus, this stage aims to: to break the matrix in order to reduce the degree of crystallinity of cellulose; increase the fraction of amorphous cellulose (which is the most suitable form for enzymes attack or chemical attack); delignification; prevent the formation of fermentation inhibiting by-products; preserve hemicelluloses (which can also be fermented) and reduce costs (SANCHEZ; CARDONA, 2008; SILVA, 2010; ZUÑIGA, 2010; SARKAR et al., 2012).

Physical pre-treatment

In physical treatment, the biomass is washed, minced, shredded and ground. This Wash removes foreign materials such as: dirt, insects' parts and sand. It then passes through the mincer to reduce size and make crushing work easer. In the pre-treatment of sugarcane bagasse, the defibrating breaks in as many sugar cane cells as possible and during grinding the sugarcane stem is divided into two fractions: the juice (rich in fermentable sugars) and the bagasse (rich in fibers) (ROSA e GARCIA, 2009).

In the steam blast method, the ground biomass is subjected to high pressure steam and temperature for a given time. Shortly thereafter, depressurization occurs to disrupt lignin, hemicellulose and cellulose bonds because of the sudden change in temperature. Finally, the material is washed to exhaustion to remove hydrolyzed sugars (SILVA, 2010).

Ultrasonic waves may be employed for pre-treatment; sound waves correspond to the region of the acoustic spectrum which is subdivided into three main regions: the infrasound range ($v < 20$ Hz); the range sounds (20 Hz$<v<$ 20 Hz) that are noticeable to humans and the ultrasound range ($v > 20$ kHz) (KORN; PEREIRA; BORGES, 2005). Ultrasound propagates through matter at speeds ranging from hundreds of meters per second in the air, to thousands of meters per second in solids (KORN; PEREIRA; BORGES, 2005; BRITO, 2011).

In addition, these waves can be generated with high and low power. low frequency ones may have high power, which causes physical and chemical modifications in the sample due to the cavitation process known as vapour bubbles. High-frequency waves have low power and are used in non-destructive tests, as well as being applied for diagnostic purposes in medicine and engineering (KORN; PEREIRA; BORGES, 2005; SORIANO, 2006; BRITO, 2011).

The thermo-hydrolysis method uses water heated at pressures above the saturation point to hydrolyze hemicellulose. biomass contact with water occurs in high pressure liquid medium and within 15 min can have temperatures ranging from 392 ºF to 446 ºF. This process can dissolve up to 60% of biomass with removal of 4% to 22% of the cellulose, 35% to 60% of lignin and all hemicellulose (GONÇALVES FILHO, 2011).

Another process involving heat is pyrolysis, where the materials are treated at temperatures above 300ºC, which results in rapid decomposition of cellulose and generates gaseous products such as H_2, CO and residual carbon. With pyrolysis, decomposition is much slower than with chemical pre-treatments (PRASAD; SINGH; JOSHI, 2007; SARKAR et al., 2012).

Chemical pre-treatment

Chemical pre-treatments are based on hydrolysis and involve dilute and concentrated acids, alkalis, wet oxidation, organic solvents and others. In this process, lignin fragmentation and solubilization of the hemicellulose fraction occur to provide greater efficiency in the cellulose hydrolysis reaction.

When concentrated acids are employed, care must be taken as to the composition of the equipment used in the process, as such products are highly corrosive and hazardous. In addition, due to economic reasons, acid recovery is required, the process of which requires considerable energy expenditure. Thus, the high investment and maintenance costs reduce the commercial interest of this process (PANOSSIAN et al., 2012).

The dilute acid method can remove almost any percentage of hemicellulose. However, it is not effective for dissolving lignin, that is, it can disrupt its bonds by increasing the susceptibility of cellulose to enzymatic hydrolysis. Breaking these bonds releases a number of compounds: sugars such as xylose, glucose and arabinose, but also oligomers, furfural and acetic acid. For this reason, care should be taken as to the amount of inhibitors generate during the reaction so that there is no low yield in alcoholic fermentation. However, cellulose is not affected due to its strong bonds.

Emmel et al., (2003) described a process employing 0.087 and 0.175% (m/m) H_2SO_4 at temperatures of 392 – 410 °F, for 2 - 5 min for pre-treatment of biomass from Eucalyptus Grandis. The best conditions for hemicellulose recovery were obtained at 410 °F for 2 min; while the lowest temperature of 392 °F, was sufficient to obtain the highest cellulose conversion yield (90%) by enzymatic hydrolysis.

According to Hamelinck et al., (2005), delignification with alkalis catalyzed hydrolysis such as NaOH or $Ca(OH)_2$ allows the base to facilitate lignin removal and increase the reactivity of the remaining polysaccharides. pre-treatment with diluted NaOH, according to Balat (2011), causes: biomass dilation, increase in internal surface area, decrease in crystallinity, separation of structural bonds between lignin and glycides and breakdown of lignin structure.

This pre-treatment removes the acetyl group and the various carboxylic acids in hemicellulose that lower the accessibility of enzymes to the surface of hemicellulose and cellulose (MOSIER et al., 2005). According to Alvira et al., (2010), catalyzed hydrolysis causes less sugar degradation than acid treatment and proves to be more effective in agricultural residues than woody materials.

Sun, Lawther and Banks (1995) studied the efficiency of different alkaline solutions through the analysis of delignification and dissolution of hemicellulose in wheat straw. They found that the ideal process condition is 1.5% NaOH in 144 hours at 68 °F, with 60% and 80% release of lignin and hemicellulose, respectively. The main disadvantage of this pre-treatment relates to some alkaline solutions which may be converted to unrecoverable salts, or may be incorporated as salts in the biomass by the pre-treatment reactions.

The organosolvent method (also known as organosolvent) uses recoverable organic solvents such as: methanol, ethanol, acetic acid, performic acid and acetone. These solvents are capable of removing lignin without the use of aggressive chemical reagents, such as caustic soda. According to Cardona, Quintero and Paz (2010), the combination of organic solvents with water eliminates the need to burn the liquor and allows the isolation of lignins.

Another advantage of organosolv, proposed by Hernandez (2007), is the volatility of the solvent used in this process, since it facilitates its recovery by distillation and reduces the environmental impact in relation to other processes. Hamelinck et al. (2005) also highlight in their work the aspect that such removal is necessary, because they can be inhibitory to the growth of microorganisms in the fermentation stage.

The fluidized bed reactor (FBR) method is a combined reaction process between hydrolysis and agitator-free fermentation that allows for excellent solids and liquid homogeneity and also reuses the lignin solution up to seven times. This procedure is economically viable because it does not use mechanical equipment with high energy demand, and can provide different applications during the process (ANTUNES et al., 2018).

Physical and Chemical Pretreatment

The integration of physical and chemical processes has gained space in academic research, especially in the pre-treatment stage. The most commonly used methods include: thermochemical treatments (such as steam explosion, or also called steam disruption); ammonium fiber burst (AFEX) (SUN; CHENG, 2002) and CO_2 burst (HAMELINCK et al., 2005).

According to Sun and Cheng (2002), the use of acid catalyzed steam explosion (H_2SO_4), increases the efficiency of enzymatic depolarization, removes hemicellulose and decreases the production of inhibitor compounds. The conditions of this sugarcane bagasse pre-treatment have been described by Morjanoff and Gray (1987) as: 428 ºF, 30 seconds of residence, 1% of H_2SO_4 followed by rapid decompression.

The method called AFEX (Ammonia Fiber Explosion) resembles steam explosion, but differs in the use of liquid ammonia as a catalyst. However, Sun and Cheng (2002) point out that the process improves hemicellulose hydrolysis and extraction, but presents inferior results in high lignin biomass.

Hamelinck et al., (2005) cite the process of CO_2 explosion, which also resembles the steam explosion. Glucose yields in subsequent enzymatic hydrolysis are low (75%) compared to steam bursts and AFEX. However, in general, CO_2 explosion costs better than ammonia explosion and does not cause the formation of inhibitors such as vapor explosion.

Biological pre-treatment

Several fungal micro-organisms have been studied in pre-treatment to improve enzymatic saccharification of lignocellulosic biomass in bioethanol production processes (ALVIRA et al., 2010). However, only a few naturally occurring fungi have the ability to degrade lignin in satisfactory proportions; these include, those belonging to the Basidiomycotina division, known as "wood degrading fungi". They are classified into three groups: white, brown and soft degrading fungi, with white-degrading fungi having higher efficiency.

Among the advantages of biological treatment, we can mention: process sustainability, low cost, low energy consumption, do not require chemical and operate under moderate pressure and temperature conditions. However, due to the long operating time, the microbial inhibition caused by degradation by-products and the low hydrolysis rate achieved in most materials (when compared to other technologies), make their diffusion and commercial scale unfeasible (ZUÑIGA, 2010).

Comparison of pre-treatment process

Table II provides an overview of advantages and disadvantages, found in the literature described in this article, on the different types of pre-treatment.

Table II – Summary of the advantages and disadvantages of some pre-treatment methods.

Pre-treatment	Advantages	Disadvantages
Physical		
Milling, dry or wet vibration.	Decreases particle size, increasing surface area. In some cases, reduces the degree of polymerization and crystallinity.	It is not sufficient to increase the sugar conversion rate. It does not remove the lignin.
Extrusion	Due process to various types of biomass; Continuous process and easily adaptable to industrial scale; Produces no effluent.	High energy consumption; Technology under development.
Irradiation	Can be combined with other pre-treatment methods.	High cost; Difficult to apply industrial.
Explosion of CO_2	Increases accessible surface area; Good value for money; There is no formation of toxic compounds.	Unaffected the lignin and hemicellulose; Requirement of high pressures.
Ultrasound	Can be combined with other types of pre-treatments.	Technology under development.
Fluized bed reactor	Allows homogeneity of the mass; Reuses the lignin solution until seven times.	Technology under development.

Chemical		
Organic solvents	Causes hydrolysis of lignin and hemicellulose.	High cost; The solvent must be drained and recycled.
Alkaline wet oxidation	Minimal training of inhibitors; Efficient removal of lignin; Exothermic process: little necessary energy in the process.	Cost of oxygen and alkaline catalyst (Na_2CO_3).
Acid hydrolysis	Concentrated acids: High performance into glucose; Room temperature. Dilute acids: Less problems with corrosion; Less inhibitor formation.	Concentrated acids High cost and e need for recovery; Equipment corrosion problems; Inhibitor formation. Dilute acids: Generation of degradation products; Low sugar concentration in the output current.
Basic hydrolysis	More effective in solubilizing lignin; Causes less degradation of sugars than acid hydrolysis.	Possible loss of fermentable sugars; Possible production of inhibitor compounds; Costs associated with effluent treatment.
Biological	Degrades lignin and hemicellulose; Low energy consumption; Soft pre-treatment (great for materials with high carbohydrate content).	Low hydrolysis rate; Greater residence time; Bacteria/fungi consume little of available carbohydrate - less sugar yield.

As shown in the table above, physical processes have advantages in being integrated with other pre-treatment methods, increasing overall process throughput. However, on another perspective of understanding, this fact can be cited as a disadvantage, because it is observed that most of the processes in this category, when not combined with other pre-treatment methods, cannot significantly affect the lignin.

In relation to chemical pre-treatments, there are the disadvantages of high costs, equipment wear and the need for better process control systems; However, there is the advantage of obtaining more effective lignin hydrolysis and better yields. Thus, as a possibility to increase productivity, making the overall process cheaper, the combined physical-chemical treatments has emerged. In the short term, this junction can yield good results, precisely because of its ability to make large-scale production of E2G feasible.

As for biological pre-treatments, greater investments are needed in research on known microorganisms (better reaction conditions); biomasses are more accessible to these microorganisms and the development, with genetic engineering, of new microorganisms. However, these systems are promising.

V. CONCLUSION

First generation ethanol is consolidated and is the main way to obtain this biofuel. However, the viability of second-generation bioethanol is of great importance to increase its availability as a fuel, making bioethanol as a clean and sustainable energy alternative. The development of E2G is beneficial to health and the environment (decreases greenhouse gas emissions) and for food generation, since cellulosic ethanol does not need to compete for space with food crops.

Given the speed with which research advances, without having a fixed or universal parameter for process qualification, the different ways in which yield is reported and which sugars are included, it is difficult to establish which process is most efficient for lignocellulosic bioethanol production. Thus, comparing the works cited comparatively is the best performed to present the results in this work.

The different forms of biomass available to generate bioethanol and the different stages of obtaining the product are broad sources of research, with the possibility of numerous and different objectives to be evaluated. To obtaining a higher yield with specific biomasses, further scientific investigations are required at the E2G pre-treatment stage. It should be considered that efficient pre-treatment processes for one type of lignocellulosic material, may not be viable for another type of biomass, making its application inefficient.

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