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ACID AND ALKALINE HYDROLYSIS TECHNOLOGIES FOR BIOETHANOL PRODUCTION: AN OVERVIEW

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ABSTRACT

In recent years, bioethanol production from renewable non-crop sources, viz., lignocellulosic biomasses has been proved a promising alternative energy source not only for compensating with the fast depleting crude oil resources, low cost, great potential availability but also for dwindling air toxics of the transportation sector. The primary technical and economic challenge for the production of bioethanol is cost- effective release of the locked polymeric carbohydrates of the highly complex lignocellulosics to fermentable soluble sugars as the bioconversion is a multistep process consisting of pretreatment, hydrolysis and fermentation. Hydrolysis of the lignocellulosic biomass can be performed either by enzymatic or acidic or alkaline methods. This paper gives an overview on the current status and up-to-date progress of the two oldest methods, acid and alkaline, of hydrolysis. Acid hydrolysis has been utilized for converting cellulosic biomass into ethanol since 1898 when the first commercial plant was established. It has been confirmed by various experimental results that alkaline hydrolysis has the highest reaction rates, followed by acid hydrolysis and finally hydrothermal degradation so far as the cleavage of glycosidic bonds in water-soluble carbohydrates is concerned. However, it is very difficult to obtain a high yield of sugar by alkaline hydrolysis because mono- and dimeric carbohydrates, such as glucose, fructose, or cellobiose, are severely attacked by alkalis at temperatures below 100° C.Albeit, both acid and alkaline hydrolyzing methods can be used for most of the lignocellulosic biomasses but as compared with the enzymatic hydrolysis the popularity of these methods is found to be diminishing concurrently owing to the low yield of sugars as well as formation of inhibitory compounds like furfurals.

Keywords: Hydrolysis, Acid Hydrolysis, Alkaline Hydrolysis, Lignocellulosic Biomass, Fermentable Sugars

I INTRODUCTION

Fuel ethanol produced from biomass (Bioethanol) seems to be a promising approach to reduce the use of non-renewable fossil fuels which are tremendously augmenting not only global warming but also environmental pollution. Therfore, the future of oil production is one of the major causes of public and private interests in developing ethanol as an additive or substitute for fossil fuel (Ramanthan 2000; Yu *et al.* 2008). This is true especially when the oil peak is estimated to reach sometime between 1996 and 2035 (Demirbas 2008). In 2010, ethanol replaced 8% of conventional gasoline globally (Brazil: 50%, US: 9%, China: 2%, EU: 1%). Thus sustainably produced biofuels can meet at least 25% of the global need for transport fuel in 2030 and potentially 50% or more going forward. Moreover, fuel ethanol can reduce green house gas emissions by 30−90% as compared to average gasoline which could also be conserved for future use. Savings by cellulosic ethanol can become even larger. Currently, the most important feedstock for the production of ethanol is sugarcane juice in Brazil and corn in the USA, while many other agricultural raw materials are also used worldwide (Rabelo *et al.* 2008; Arumugam & Manikandan 2011). The cost of ethanol production increases as the demand for molasses increase. Hence, non-crop lignocellulosic biomasses such as wood, switch grass, aquatic weeds and agricultural residues are attractive materials for the ethanol production since they are the most abundant reproducible resources on earth (Saha *et al.* 2005; Jefries 2006; Sun & Chen 2007; Mishima *et al.* 2008; Cheng & Keshwani, 2008; Ying *et al.* 2009). Agricultural residues are easier than wood to use as feedstocks for biofuels due to their lower lignin and higher hemicellulosic contents. Therefore, there is a hope that large production of ethanol from lignocellulosic biomass resource becomes a reality by 2015 (Seabra *et al.* 2010).

The main components of lignocellulosic biomass are cellulose, hemicelluloses and lignin. The most abundant polysaccharide on earth, cellulose, is a highly ordered polymer of cellobiose representing over 50% of the wood mass (Bon *et al.* 2007). Both lignin and cellulose are more recalcitrant to enzymatic or chemical conversion to biofuel precursors. Hemicellulose is much more readily recovered, but presents challenges in use due to simultaneous release of acetic and ferulic acid components along with acid-soluble lignin moieties, all of which can inhibit fermentation and catalytic conversion to more useful products (Oliva *et al.* 2005; Alvira *et al.* 2010; Marinkovic *et al.* 2012). Hence the primary technical and economic challenge for the production of bioethanol is cost effective release of the locked polymeric carbohydrates of the highly complex lignocellulosics to fermentable soluble sugars as the bioconversion process consists of at least four main steps: pretreatment, hydrolysis, fermentation and recovery and purification (Sun & Cheng 2002; Geng *et al.* 2009; Kuhad *et al.* 2010). To produce ethanol commercially profitable from the lignocellulosic biomass (LB), there is a need for technological improvement and cost reduction in all the stages of production (Hamelinck *et al.* 2005; Rabelo *et al.* 2008; Gupta *et al.* 2009; Payamera *et al.* 2011; Phuengjayaem *et al.* 2011). The pretreatment stage has been found to be the key step to provide a substrate susceptible to the subsequent hydrolysis (Wyman *et al.* 2005; Mosier *et al.* 2005a,b; Binod *et al.* 2010; Harun *et al.* 2011). Hydrolysis is the process of breakdown of cellulose into cellobiose and glucose which can be accomplished either by enzymes or acid or alkali (Xiang *et al.* 2004; Taherzadeh & Karimi 2008). Of these methods, dilute acid hydrolysis and enzymatic hydrolysis have been the most popular ones. Dilute acid hydrolysis is a fast and convenient method to perform but it leads to the accumulation of fermentation inhibitory compounds such as carboxylic acids, fufurals, hydroxyl methyl furfurals (HMF) and phenolics (Gupta *et al.* 2009; Mukherjee *et al.* 2010). However, the cost of ethanol production from lignocellulosic biomass is relatively high based on current technologies. Acid hydrolysis is perhaps currently seen as the most technologically mature method of sugars release from biomass. The concentrated acid process uses relatively mild temperatures, and the only pressures involved are those created by pumping materials from vessel to vessel. Alkaline hydrolysis is also considered to be more suitable for herbaceous crops and agricultural residues (Huber *et al.* 2006)**.** The aim of this review is the conversion of LB into monomeric sugars with the help of acid or alkaline hydrolysis so that the released monomeric sugars could be used for fermentation to ethanol easily either by using *Saccharomyces cerevisiae* or *Picchia stipitis*.

1.1 Acid Hydrolysis

Acid hydrolysis is the oldest technology for converting cellulose biomass to bioethanol (Graf *et al.* 2000). There are two basic types of acid hydrolysis processes commonly used: dilute acid and concentrated acid hydrolysis. The dilute acid process is conducted under high temperature and pressure and has a reaction time at a scale of up to minutes, facilitating continuous processing. The concentrated acid process uses relatively mild conditions with a much longer reaction time (Sun & Cheng 2002, 2007; Mishima *et al.* 2006, 2008; Masami *et al.* 2008).

1.2 Dilute Acid Hydrolysis

The main step is essentially hemicellulose fraction is depolymerized at lower temperature than the cellulosic fraction. Dilute sulfuric acid is mixed with biomass and held at temperatures of 160-220 °C for periods ranging from minutes to seconds to hydrolyze hemicellulose to xylose and other sugars, and then continue to break xylose down to furfural (Balat *et al.* 2007). The dilute acid process involves a solution of about 1% sulfuric acid concentration in a continuous-flow reactor at a high temperature (215°C) (Graf *et al.* 2000). Most dilute acid processes are limited to a sugar recovery efficiency of around 50%. The primary challenge for dilute acid hydrolysis processes is how to raise glucose yields higher than 70% in an economically viable industrial process while maintaining a high cellulose hydrolysis rate and minimizing glucose decomposition. Percolation reactors have been used in most of the wood sugar processes (Xiang *et al.* 2004). Countercurrent shrinking bed reactor technologies have been successful in achieving >90% glucose yield from cellulose. Anwar *et al.* 2012 studied dilute acid hydrolysis of rice polish influenced on releasing hemicelluloses to produce xylose, arabinose, glucose, acid-soluble lignin, but there was also released of some inhibitory compound like furfural and phenolic compound which affect the hydrolysis process. They reported that hydrolysis with H_2SO_4 can be carried out at elevated temperatures (80-200 ºC) for 2 – 60 min. They also found that H_2SO_4 is more efficient as a catalyst than hydrochloric acid (HCl) for the degradation of xylose. Saha *et al.* (2006) described a process for the hydrolysis and conversion of rice-hull cellulose and hemicellulose to monomeric sugars. They used dilute acid H_2SO_4 pretreatment at varied temperatures and enzymatic saccharification. Maximum yield of monomeric sugars by dilute acid pretreatment and enzymatic saccharification using commercial cellulases was 60% based on total carbohydrate content. Cheng *et al.* (2008) evaluated the cellulose reactivity of two lignocellulosic feedstocks, switch grass and poplar, using dilute sulfuric acid pretreatments designed for optimum xylose yield. Yields (percentage conversion of cellulose) were 90% and 73% of the theoretical yield for pretreated switch grass and poplar, respectively (Balat *et al.* 2007). Saha & Cotta (2006) used dilute acid and enzymatic saccharification procedures for conversion of corn fiber to fermentable sugars. They found that corn fiber pretreated with 0.5% H_2SO_4 at 121ºC for 1 h facilitated commercial enzymes to highly hydrolyze remaining starch and hemicellulose components without generation of inhibitors such as furfural and hydroxymethyl furfural (HMF), which are generally considered inhibitors for fermentative microorganisms. The acid must be neutralized before the sugars proceed to fermentation (Kim *et al.* 2012). Acid hydrolysis releases oligomers and monosaccharides and has historically been modeled as a homogeneous reaction in which acid catalyzes breakdown of cellulose to glucose followed by breakdown of the glucose released to form HMF and other degradation compounds (Balat *et al.* 2007; Hsu *et al.* 2010). Strong acids can reduce the crystalline region but they degrade glucose. The sulfuric acid-based hydrolysis process is operated under two different conditions; (í) a process that uses high sulfuric acid concentration that operates at a lower temperature and, (íí) a process that uses low sulfuric acid concentration and operates at a higher temperature stage is optimized for hydrolysis of the cellulose portion of the feedstock (Kumar *et al.* 2005; Kim & Mazza 2009). The first process is conducted under mild process conditions to recover the 5-carbon sugars while the second stage is conducted under harsher conditions to recover the 6-carbon sugars (Dermirbas 2005, 2006, 2007). Accordingly, in the first stage hemicellulose is hydrolyzed (Figure 1) with dilute acid under ambient conditions and the more resistant cellulose is hydrolyzed at higher temperatures (213°C) and relatively higher concentration of acid (0.4%) in the second stage. Under the latter conditions, the recovery yields are 89% for mannose, 82% for galactose and 50% for glucose (Graf & Koehler 2000; USDOE 2003). Schematic flowsheet for dilute acid hydrolysis is given in

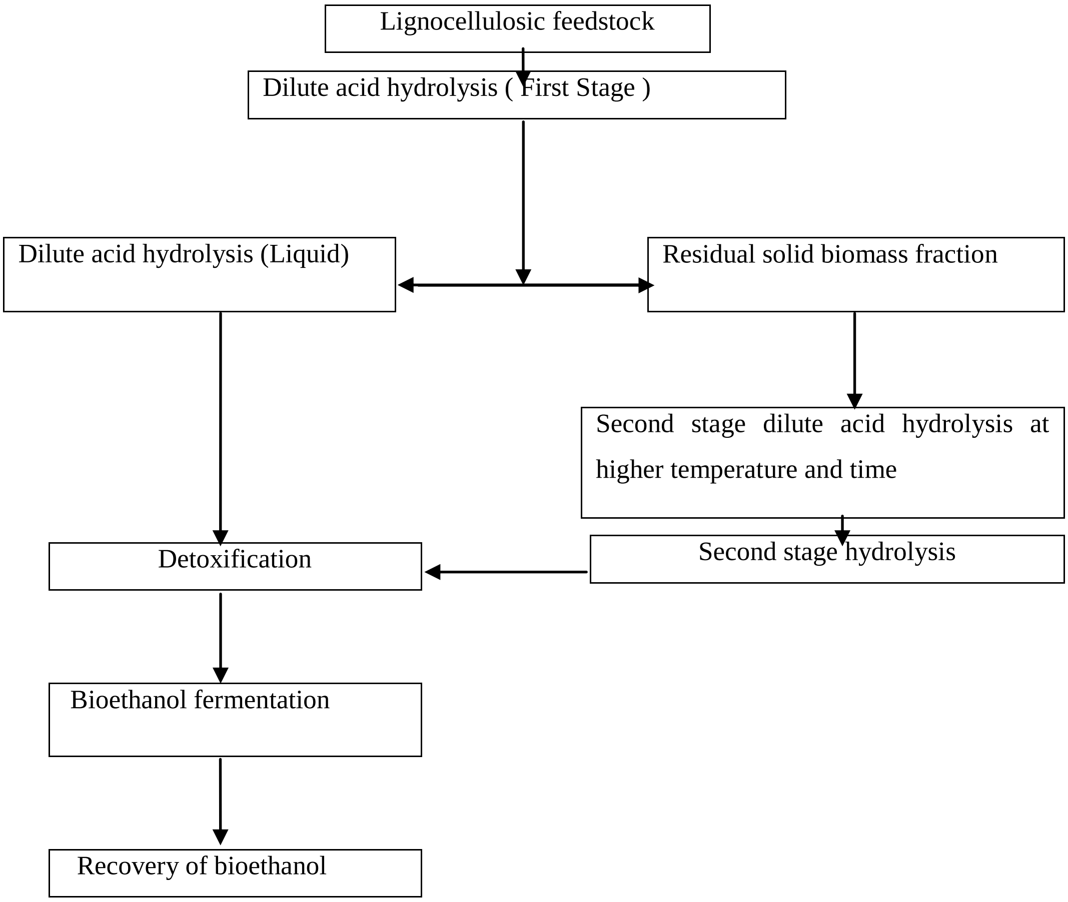

Figure 1. Dilute acid hydrolysis (first stage and two stage) and recovery of bioethanol.

Figure. 1. In contrast, the concentrated acid methods result in high sugar recovery yields (90%), can handle diverse feedstocks containing lignocellulose and the process is relatively rapid (10 to12 h), (Joshi *et al.* 2011). The acid hydrolyzed substrates were then subjected to enzyme hydrolysis to give vastly improved yields as high as 100% for corn stover and 90% for oak wood (Balat *et al.* 2007). The advantage of dilute acid processes is the combination of higher conversion and cheaper utility costs, their fast rate of reaction, which facilitates continuous processing. The biggest limitation is their low sugar yield and corrosion, which mandates expensive materials of construction. The acid must be neutralized before the sugars proceed to fermentation. The dilute acid process involves a solution of about 1-2% sulfuric acid concentration at a high temperature. Nutawan Yoswathana *et al.* (2010) have utilized dilute sulfuric acid (1-9%) for the hydrolysis of rice straw for ethanol production. The major advantage of dilute acid hydrolysis is that it is quicker than concentrated acid hydrolysis and hence can be used as a continuous process. The disadvantage of this method is that the sugar conversion efficiency is only about 50%. Also due to high temperature and pressure, large portion of the sugars is degraded which is not fermentable. The reducing yield of water hyacinth (366.0 mg/g dry matter) for the acid and combined hydrolysis at 100°C for 60 min (Ma *et al.* 2010).

The highest sugar yield on water hyacinth (155.13 mg/sugar/g dry matter) obtained by combination of drying, grinding and hydrolysis with 5% (Harun *et al.* 2011). The maximum yields of glucose from sweet sorghum straw were 0.234 g/g dry substrate, respectively, at the pretreatment condition: 120 ºC, 3% H_2SO_4 for 10 min (Phuengjayaem &Teeradakorm 2011).

Table 1. **Total Sugar yield under different hydrolysis conditions for different feedstocks**

S. No	**Feedstock**	**Hydrolysis conditions**	**Sugar yield**	**Reference**
1.	Cassava waste	121°C, pH 4.5, 24 h	6.2%	Srinorakutara *et al.* (2004)
2.	Wheat straw	45°C, pH 5.0, 72 h	56.5 %	Saha *et al.*(2005)
3	Wheat straw	45°C, pH5.0, 120 h	67.2%	Saha *et al.* (2006)
4.	Wheat straw	50°C, pH 4.8, 24 h	51.4 %	Tabka *et al.* (2006)
5.	Water hyacinth	37°C, pH 5.0, 24 hr	19.90%	Mishima *et al.* (2006)
6.	*Garcinia kola*	50°C, pH 5.0, 144 h	86.2%	Humphray *et al.* (2007)
7.	Sugarcane bagasse	70°C, pH 5.0, 6 h	62.4%	Rabelo *et al.* (2008)
8.	Olive tree	50°C, pH 4.8, 72 h	36.3%	Cara *et al.* (2008)
9.	Switch grass	50°C, pH 4.8	58.7%	Yu *et al.* (2008)
10.	Switch grass	50°C, pH 4.8	58.7%	Hu *et al.* (2008)
11.	Agave bagasse	121°C, pH 4.8, 4 h	12-58%	Hernandez-Salas *et al.* (2009)

10.	Sugarcane bagasse	121°C, pH 4.8, 4 h	13-18%	Hernandez-Salas *et al.* (2009)
11.	*P. juliflora* wood	45°C, pH 5.0, 120 h	67.2%	Gupta *et al.* (2009)
12.	Bamboo	120-140°C, pH 5.0,90 min	8.5%	Payamera *et al.* (2011)
13.	Water hyacinth	121°C, pH 4.8, 1 h	35.9%	Harun *et al.* (2011)
14.	Cassava pulp	210°C, 15 min	52.27%	Hermiati *et al.* (2012)

1.3 Concentrated Acid Hydrolysis

This method provides a complete and rapid hydrolysis of cellulose to glucose and hemicelluloses to 5-carbon sugars with little degradation. The critical factors needed to make this process economically viable are to optimize sugar recovery and cost effectively recovering the acid for recycling (Dermirabas 2004, 2005). The concentrated acid process uses relatively mild temperatures, and the only pressures involved are those created by pumping materials from vessel to vessel. Reaction times are typically much longer than for dilute acid process (Graf *et al.* 2000). The concentrated acid process uses 70% sulfuric acid at 313–323K for 2–4 h in a reactor. The low temperatures and pressures will lead to minimization of the sugar degradation. In the next step, the cellulosic fraction has to be deploymerized. The solid residue from first stage is de-watered and soaked in 30–40% sulfuric acid for 50 min. at 373K for further cellulose hydrolysis (Chandel *et al.* 2007). The primary advantage of the concentrated acid process is the potential for high sugar recovery efficiency (Dermirbas 2005). The concentrated acid process offers more potential for cost reductions than the dilute sulfuric acid process (Farooqi *et al.* 2004). Concentrated sulfuric or hydrochloric acid is difficult to work with, and essentially all of the acid must be recovered and reconcentrated in order for the process to be economical. Conventional dilute acid cellulose hydrolysis has been unpopular because sugars decompose under conditions that are required for cellulose hydrolysis, i.e., high temperature and low pH. Therfore, according to Ma *et al.* (2011) the optimum hydrolysis conditions are as follows: the concentration of H_2SO_4 is 72% (wt.%), the temperature is 50°C, the ratio of H_2SO_4 solution volume (mL) to the rice hull mass (g) is 10:1 and the time is 5 minutes, the glucose yield rate reaches 45.6% (wt.%).

1.4 Alkaline Hydrolysis

Alkaline hydrolysis (mainly lime (CaO/Ca(OH)2), NaOH, Na_2C0_3) is effective in removing lignin and acetyl groups (Mohan *et al.* 2006; Yang & Wyman 2007). Alkaline hydrolysis is a slow process, requires neutralization, and the added alkali also needs to be recovered. Alkaline hydrolysis is more suitable for herbaceous crops and agricultural residues and not suited for woody biomass due to its higher lignin content (Huber *et al.* 2006; Rabelo *et al.* 2008). The OH^-ion attacks the anomeric carbon atom and cleaves the ether bridge during alkaline hydrolysis. With the uptake of water and liberation of the OH^- ion, glucoses are formed. It has been confirmed by various experimental results that alkaline hydrolysis has the highest reaction rates, followed by acid hydrolysis and finally hydrothermal degradation so far as the cleavage of glycosidic bonds in water-soluble carbohydrates is concerned. However, to obtain a high yield of sugar by alkaline hydrolysis, it is very difficult because mono- and dimeric carbohydrates, such as glucose, fructose, or cellobiose, are severely attacked by alkalis at temperatures below 100 °C ((Wyman *et al.* 2005; Rabelo *et al.* 2008). During hydrolysis, organic acids are also formed therefore, the alkali consumption by acid formation is also a problem. Alkaline hydrolysis can be used for the pretreatment of lignocellulosic biomass, being saponification of intermolecular ester bonds cross-linking xylan hemicellulose and other components, e.g., lignin and other hemicelluloses (Sun *et al*. 2002; Yang & Wyman 2007). Dilute NaOH treatment of lignocellulosic biomass causes swelling, leading to an increase in the internal surface area, a decrease in crystallinity, separation of structural linkages between lignin and carbohydrates, and disruption of the lignin structure (Fang *et al.* 1987; Kim & Lee 2007; Yu *et al.* 2008; Kumar & Wyman 2009). The yield of total sugars of 672+ 4 mg per g of wheat straw hydrolysis with alkaline H_2O_2 (2.15%, v/v; pH 11.5; 35 °C; 24 h) after enzymatic saccharification (45 °C, pH 5.0, 120 h) using 0.16 mL of enzyme per g of straw (Saha & Cotta 2006). The yield of total sugars of 494.7 mg per g of sugarcane bagasse , the best results are for alkaline peroxide hydrolysis performed with 5% H_2O_2 at 20°C for 24 h (Rabelo *et al.* 2008). The monosaccharide yield was about 80% at 35°C for 10% alkali concentration in a reaction time of 90 minutes (Geeta & Gopalakrishnan 2011).

II COMPARISON OF DIFFERENT HYDROLYSIS METHODS

In comparison to dilute acid hydrolysis, concentrated acid hydrolysis leads to little sugar degradation and gives sugar yields approaching 100%. However, environmental and corrosion problems and the high cost of acid consumption and recovery present major barriers to economic success. The major advantage of dilute acid hydrolysis is that it is quicker than concentrated acid hydrolysis and hence can be used as a continuous process. Mild acid hydrolysis is a promising method to improve enzymatic hydrolysis and ethanol production. At high temperature (70 and 100°C), the reducing sugar yield increased with the increase in hydrolysis time (Yu *et al.* 2008; Ma *et al.* 2010). Table 2 is also drawn below to depict the comparisons of various hydrolysis methods including enzymatic hydrolysis which seems to be the future method of hydrolysis of various lignocellulosic biomasses for obtaining monomeric sugars which could be fermented into boehtanol.

Table 2: Comparisons of different hydrolysis methods

S. No.	Hydrolysis method	Conditions	Glucose yield (%)	Advantages	Disadvantages
1	Dilute acid	<1% H_2SO_4, 215°C, 3 min	50–70	• High sugar recovery • Very high reaction rate	• Environmental and corrosion problems • Sugar decomposition at elevated temperature • High utility cost for elevated temperature • High operating cost for acid consumption
2	Concentrated acid	30–70% H_2SO_4, 40°C,	90	• High sugar recovery • High reaction rate	• Environmental and corrosion problems • High cost for acid

		2–6 h			recovery
3	Alkaline	18% NaOH 100°C, 1 h	30	High reaction rate	• Low sugar yield • Sugar decomposition by alkali attack
4	Enzymatic	Cellulase 70°C, 1.5 days	75 -95	• High yield of relatively pure sugar • Mild environmental conditions • No environmental and corrosion problems	• Pretreatment of biomass required • High cost of cellulase enzymes • Low hydrolysis rate

III CONCLUSION

Lignocellulosic biomass materials are cheap feedstocks for ethanol production. The key challenge is to develop hydrolysis technologies that are capable of recovering sugars effectively and efficiently (Geetha *et al.* 2011). One major problem with ethanol production is the availability of raw materials which can vary considerably from season to season and depend on geographic locations. The price of the raw materials is also highly volatile which can highly affect the production costs of bioethanol. Because feedstocks typically account for greater than one-third of the production costs, maximizing bioethanol yield is imperative. Dilute acid hydrolysis achieves a sugar yield of 50–70%. and alkaline hydrolysis achieves a low sugar yield of 30%. At present, enzymatic hydrolysis can obtain a sugar yield of 75–95% and further research project yield of 85–95%. Although acid hydrolysis processes are matured technologies, enzymatic processes have comparable costs and the potential of future cost reductions as technology improves. Future research is needed to develop suitable catalysts to facilitate cellulose hydrolysis at relatively low temperatures to minimize the decomposition of sugar products. Although bioethanol production has been greatly improved by new technologies, several research challenges remain in order to further improve the overall yield of ethanol, increase the productivity in the conversion steps and to reduce the production cost. These challenges include maintaining a stable performance of the genetically engineered yeasts in commercial scale fermentation operations, developing more efficient pretreatment technologies for lignocellulosic biomass, improving fermentation of all sugars available in wood and to make the fermenting organism more tolerant to inhibitors, increasing process integration to reduce the number of process steps, the energy demand and to re-use process streams in order to minimize the use of fresh water and reduce the amount of waste streams.

REFRENCES

[1.] Anwar, Z., Gulfraz, M., Asad, M.J., Imaran, M., Akram, Z., Mohmood, S., Rehman, A.,Anwar, P. & Sadiq, A. 2011 Bioethanol productions from rice polish by optimization of dilute acid pretreatment and enzymatic hydrolysis. *African Journal of Biotechnology* 11, 992-998.

[2.] Alvira, P., Tomas-Pejo, P., Ballesteros, M. & Negro, M.J. 2010 Pretreatment technologies for an efficient bioethanol production based on enzymatic hydrolysis: A review. *Bioresource Technol* 101, 4851-4861.

[3.] Arumugam, R. & Manikandan, M. 2011 Fermentation of pretreated hydrolyzates of banana and mango fruit wastes for ethanol production. *Asian Journal Exp.Biological Sciene* 2, 246-256.

[4.] Balat, M., Balat, H. & Cahide, O. 2007 Progress in bioethanol processing. *Progress Energy Combust Science* 34, 1-23.

[5.] Binod, P., Sindhu, R., Singhania, R., Vikram,S., Devi, L., Nagalakshmi, S., Kurien, N., Sukumaran, R. & Pandey, A. 2010 Bioethanol production from rice straw: an overview. *Bioresource Technology* 101, 4767-4774.

[6.] Bon, E.P.S. & Ferrara, M.A. 2007 Bioethanol production via enzymatic hydrolysis of cellulosic biomass, *An FAO Seminar on The Role of Agricultural Biotechnologies for Production of Bioenergy in Developing Countries, Rome.*

[7.] Cara, C., Moya, M., Ballesteros, I., Negro M.J., González, A. & Ruiz, E. 2007 Influence of solid loading on enzymatic hydrolysis of steam exploded or liquid hot water pretreated olive tree biomass. *Process Biochemistry* 42, 1003-1009.

[8.] Cara, C., Ruiz, E., Ballesteros, M., Manzanares, P., José Negro, M. & Castro. 2008 Production of fuel ethanol from steam-explosion pretreated olive tree pruning. *Fuel* 87, 692-700.

[9.] Chandel, A.K., Chan, E.S., Rudravaram, R., Narasu, M.L., Rao, L.V. & Ravindra, P. 2007 Economics and environmental impact of bioethanol production technologies: an appraisal. *Biotechnol Mol Bio Rev* 2, 14–32.

[10.] Cheng, K.-K., Cai, B.-Y., Zhang, J.-A., Ling, H.-Z., Zhou, Y.-J., Ge, J.-P. & Xu, J.-M. 2008 Sugarcane bagasses hemicellulose hydrolysate for ethanol production by acid recovery process. *Biochemical Engineering Journal* 38, 105–109.

[11.] Keshwani DR, Cheng JJ. 2008. Modeling of microwave based chemical pretreatment of switchgrass. *ASABE Annual Meeting Paper No. 083954, Providence, RI.*

[12.] Demirbas, A. 2004 Ethanol from cellulosic biomass resources. *Int J Green Energy* 1,79–87.

[13.] Demirbas, A. 2005 Estimating of structural composition of wood and non-wood biomass samples. *Energy Source* 27, 761–7.

[14.] Demirbas, A. 2006 Hazardous emissions, global climate change and environmental precautions. *Energy Sources B* 1, 75–84.

[15.] Demirbas, A. 2008 Biofuels sources, biofuel policy, biofuel economy and global biofuel projections. *Energy Conversion and Management* 49, 2106-2116.

[16.] Demirbas AH, Dermirbas I .2007 Importance of rural bioenergy for developing countries. *Energy Conversion Management* 48, 2386-2398.

[17.] Fang, L.T., Gharpuray, M.M. & Lee, Y.H. 1987 *Cellulose Hydrolysis Biotechnology Monographs*; Springer: Berlin, Germany 55.

[18.] Farooqi, R. Sam, A.G.2004 Ethanol as a transportation fuel. *Centre for Applied Business Research in Energy and the Environment (CABREE) Climate Change Initiative.* University of Alberta, Canada.[cited; available from: /www.business.ualberta.ca/ cabreeS].

[19.] Geehta, G.S. & Gopalakrishnan, A.N. 2011 Bioethanol production from paper fibre residue using diluted alkali hydrolysis and the fermentation process .*E-Journal of Chemistry* 8, 1951-1957.

[20.] Geng, A., Xin, F. & Ip, Jun-yu. 2009 Ethanol production from horticultural waste treated by a modified organosolve method. *Bioresource Technology* 104, 715-721.

[21.] Graf, A. & Koehler, T. 2000 Oregon cellulose-ethanol study: an evaluation of the potential for ethanol production in Oregon using cellulose based feedstocks. *Salem, Oregon, USA: Oregon Dept of Energy.* 1-36.

[22.] Gupta, R., Sharma, K.K. & Kuhad, R.C .2009 Separate hydrolysis and fermentation (SHF) of *Prosopis juliflora*, woody substrate for the production of cellulosic ethanol by *Saccharomyces cerevisiae* and *Pichia stipitis* NCIM 3498. *Bioresource Technology* 100, 1214-1220

[23.] Hamelinck, C.N., Hooijdonk, G. & Faaij, A.P.C. 2005 Ethanol from lignocellulosic biomass: techno-economic performance in short-, middle- and long term. *Biomass Bioenergy* 28, 384–410.

[24.] Harun, M.Y.,Raiah, A.B.D., Abidin, Z.Z. & Yunus, R. 2011 Efecctof physical pretreatment on dilute acid hydrolysis of water hyacinth (*Eichhornia crassipes*).*Bioresource Technology* 102, 5193-5199.

[25.] Hermiati, E., Azuma, J., Tsubaki, S., Mangunwidjaja. D., Sunarti, T.C., Suparno, O. & Prasetya, B. 2012 Improvement of microwave-assisted hydrolysis of cassava pulp and tapioca flour by addition of activated carbon. *Carbohydr. Polym.* 87, 939-942.

[26.] Hernández-Salas, J.M., Villa-Ramírez, M.S., Veloz-Rendón, J.S., Rivera-Hernández, K.N., González-César, R.A., Plascencia-Espinosa, M.A., Trejo-Estrada, S.R., 2009. Comparative hydrolysis and fermentation of sugarcane and agave bagasse. *Bioresource Technology* 100, 1238–1245.

[27.] Hsu, T.C., Guo, G.L., Chen, W.H. & Hwang, W.S. 2010 Effect of dilute acid pretreatment of rice straw on structural properties and enzymatic hydrolysis. *Bioresource Technology* 101, 4907-4913.

[28.] Huber G.W., Iborra S., Corma A. 2006 Synthesis of transportation fuels from biomass: chemistry, Catalysts and Engineering. *Chemitry Rev*eiw 106, 4044-4098.

[29.] Humphery, C. N. & Okafoagu, C.U. 2007 Optimization of ethanol production from *Garcinia kola* pulp agrowaste. *African J. of Biotech.,* 6, 2033-2037.

[30.] Hu, Z *et al. (*2008)

[31.] Jeffries, T.W. 2006 Engineering yeasts for xylose metabolism. *Curr. Opin. Biotechnology* 17, 320-326.

[32.] Joshi, B., Bhatt, M.R., Sharma, D., Joshi, J., Malla, R. and Sreerama, L. 2011 Lignocellulosic ethanol production: Current practices and recent developments. *Biotechnology and Molecular Biology Review* 6, 172-182.

[33.] Kim, T.H. & Lee, Y.Y. 2007 Pretreatment of corn stover by soaking in aqueous ammonia at moderate temperature. *Appl Biochem Biotechnol* 136, 81-92.

[34.] Kim, JW. & Mazza, G. 2008 Optimization of phosphoric acid catalyzed fractionation and enzymatic digestibility of flax shives. *Ind. Crops Prod*. 28, 346-355.

[35.] Kim, J. & Mazza, G. 2009 Extraction and separation of carbohydrates and phenolics compounds in flax shives with pH controlled pressurized low polarity water. *J. Agric. Food Chem. 57*, 1805–1813.

[36.] Kim, J.W., Kim, K.S., Lee, J.S., Park, S.M., Cho, H.Y., Park, J.C. & Kim, J.S. 2011 Two –stage pretreatment of rice straw using aqueous ammonia and dilute acid. *Bioresource Technology* 102, 8992-8999.

[37.] Kuhad, R.C., Gupta, R., Khasa, Y.P. & Singh, A. 2010 Bioethanol production from *Lantana camara* (red sage): Pretreatment, saccharification and fermentation. *Bioresource Technology* 101, 8348-8354.

[38.] Kumar, B.M., Rajesh, G., & Sudheesh, K.G. 2005. Aboveground biomass production and nutrient uptake of thorny bamboo [*Bambusa bambos* (L.) Voss] in the homegardens of Thrissur, Kerala. *J. Trop. Agric.* 43 , 51–56.

[39.] Kumar, R. & Wyman, C.E. 2009 Effect of xylanse supplementation of cellulose on digestion of corn stover solids prepared by leading pretreatment technologies. *Bioresource Technoogy*. 100, 4203- 13.

[40.] Ma, F.,Yang, Y., Xu, C., Yu, H., Wu, J.& Zhang, X. 2010 Combination of biological pretreatment with mild acid pretreatment for enzymatic hydrolysis and ethanol production from water hyacinth. *Bioresource Technology* 101, 9600-9604.

[41.] Ma, Y., Zhao, X., Zhang, H., Wang, Z. 2011 Comprehensive utilization of the hydrolyzed productions from rice hull. *Industrial Crops and Products* 33, 403–408.

[42.] Marinkovic, S., Bras, J.L., Rataj, V.N., Agach, M. & Estrine, B. 2012 Acidic pretreatment of wheat straw in decanol for the production of surfactant, lignin and glucose. *International Journal of Molecular Science* 13, 348-357.

[43.] Masami, G.O.O., Usui, I.Y. & Urano, N. 2008 Ethanol production from water hyacinth *Eichhornia crassipes* by yeast isolated from various hydrospheres. *African Journal of Microbiogy Research* 2, 110-113.

[44.] Mishima,D., Tateda, M. & Fujita, M. 2008 Comparative study on chemical pretreatments to accelerate enzymatic hydrolysis of aquatic macrophyte biomass used in water purification processes. *Bioresource Technology* 97, 2166-2172.

[45.] Mishima, D., Kuniki, M., Sei, K., Soda, S., Ike, M. & Fujita, M. 2008 Ethanol production from candidate energy crops: water hyacinth *(Eichhornia crassipes*) and water lettuce (*Postia stratiotes L.*). *Bioresource Technology* 99, 2495-2500.

[46.] Mohan, D., Pittman, C.U. & Steele, P.H. 2006 Pyrolysis of wood/biomass for bio-oil: A critical review. *Energy & Fuels* 20, 848-889.

[47.] Mosier, N., Hendrickson, R., Ho, N., Sedlak, M. & Ladisch, M. 2005a Optimization of pH controlled liquid hot water pretreatment of corn stover. *Bioresourc Technology* 96, 1986-1993.

[48.] Mosier, N., Wyman, C.E., Dale, B.D., Elander, R.T., Lee, Y.Y., Holtzapple, M. & Ladisch, C.M. 2005b Features of promising technologies for pretreatment of lignocellulosic biomass. *Bioresource Technology* 96, 673- 686.

[49.] Mukherjee, S., Sen, R., Balendu, Giri., Satpute, D., Chakrabarti, T. & Pandey, R.A. 2010 Commercializing lignocellulosic bioethanol: technology bottlenecks and possible remedies. *Biofuels, Bioprod. Bioref* 4, 77–93.

[50.] Nutawan, Yoswathana., Phattayawadee, Phuriphipat., Pattranit, Treyawutthiwat. & Mohammad, N. E. 2010 Bioethanol production from rice straw. *Energy Research Journal* 1 , 26-31.

[51.] Oliveria, M.E.D., Vaughan, B.E. & Rykiel Jr.E.J. 2005 Ethanol as fuel: energy, carbon dioxide balances, and ecological footprint. *BioScience*. 55, 593–602.

[52.] Payamara, J. & Payamera, A. 2011 Fuel ethanol production from biomass conversion in Iran. *Int. Journal of Chem Tech Research* 3, 1446-1449.

[53.] Phuengjayaem, S. & Teeradakorm, S. 2011 Saccharification of acid –pretreatment sweet sorghum straw by cellulose for bioethanol production. *International Conference on Asia Agriculture and Animal IPCBEE 13,* 27-3.

[54.] Rabelo, S.C., Filho, R.M. & Costa, A.C. 2008 A comparison between lime and alkaline hydrogen peroxide pretreatments of sugarcane bagasse for ethanol production. *Appl Biochem Biotechnol* 144, 87-100.

[55.] Raghavendra, B., Havannavar, Geeta., S.G. 2007 Pretreatment of agroresidues for release of maximum reducing sugar. *Karnataka J. Agric. Science* 20, 771-772.

[56.] Ramanathan, M. 2000, Biochemical conversion ethanol production from root crops. In: *Biomass Conversion Technologies for Agriculture and Allied Industries* 157-162.

[57.] Saha, B.C., Iten,L.B., Coota, M.A. & Wu, Y.V. 2005 Dilute acid pretreatment , enzymatic saccharificatin, and fermentation of wheat straw to ethanol. *Proc. Biochem*. 40, 3693-3700.

[58.] Saha, B. C., & Cotta, M.A. 2006 Ethanol production from alkaline peroxide pretreated enzymatically saccharified wheat straw. *Biotechnology Prog* 22, 449-453.

[59.] Saha, B.C., Iten, L.B., Coota, M.A. & Wu, Y.V. 2006 Dilute acid pretreatment , enzymatic saccharificatin, and fermentation of rice hulls to ethanol. *Biotechnology Prog.* 21, 816-822.

[60.] Seabra, J., Tao,L., Chum, H. & Macedo, I. 2010 “A techno-economic evaluationof the effects of centralized cellulosic ethanol and co-products refinery options with sugarcane mill clustering”. *Biomass Bioenerg* 34, 1065-78.

[61.] Srinorakutara, T., Kaewvimol, L. & Saengow, L. 2006 Approach of cassava waste pretreatments for fuel ethanol production in Thailand. *J. Sci. Res. Chula Univ* 3, 77-84.

[62.] Sun, Y. & Cheng, J. 2002 Hydrolysis of lignocellulosic materials for ethanol production: a review. *Bioresource Technol*ogy 83, 1–11.

[63.] Sun, J.X., Sun, X.F., Zhao, H., Sun, R.C., 2004 Isolation and characterization of cellulose from sugarcane bagasse. Polym. Degrad. Stab. 84, 331–339.

[64.] Sun, F.B. & Chen, H.Z. 2007 Evaluation of enzymatic hydrolysis of wheat straw pretreated by atmospheric glycerol autocatalysis. *J. Chem. Technol. Biotechnology* 82, 1039-1044.

[65.] Tabka, M. G., Herpoël-Gimbert, I., Monod, F., Asther, M., Sigoillot, J.C. 2006 Enzymatic saccharification of wheat straw for bioethanol production by combined cellulose xylanase and feruloyl esterase treatment. *Enzyme and microbial technology* 39, 897-902.

[66.] Taherzadeh, M.J., Karimi, K. 2008 Pretreatment of lignocellulosic wastes to improve ethanol and biogas production: A review. *Int. J. Mol. Science* 9, 1621-1651.

[67.] Wyman, C.E., Dale, B.E., Elander, R.T., Holtzapple,M., Ladisch, M.R. & Lee, Y.Y. 2005 Comparative sugar recovery data laboratory scale application of lead pretreatment technologies to corn stover. *Bioresource Technology* 96, 2026- 2032.

[68.] Xiang, Q., Lee, Y.Y., Pettersson, O. P., Robert, W. Torget 2003 Heterogeneous aspects of acid hydrolysis of α-cellulose. *Applied Biochemistry and Biotechnology* 10, 505-514.

[69.] Yang, B. & Wyman, C.E. 2008 Pretreatment : the key to unlocking low-cost cellulosic ethanol. *Biofuels Bioproduction Bioref*. 2, 26-40.

[70.] Ying, Y., Shivappa, R.S., Burns, J.C. & Cheng, J.J. 2009 Dilute acid pretreatment of oven-dried switchgrass germplasms of bioethanol production. *Energy Fuels* 23, 3759-3766.

[71.] Yu, Y., Lou, X., Wu, H. 2008 Some recent advance in hydrolysis of biomass in hot-compressed water and its comparisons with other hydrolysis methods. *Energy and Fuels* 22, 46-60.