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Conversion of Lignocellulosic Biomass to Fuel Ethanol - A Brief Review

LIM KOON ONG

Bioenergy Laboratory, School of Physics, Universiti Sains Malaysia, 11800 Penang, Malaysia

The paper provides a brief review of the conversion of lignocellulose biomass to fuel ethanol. The various steps in the conversion process are discussed and some information on the potential of the conversion process are also provided.

Keywords: *Biomass, lignocellulose, ethanol, acid hydrolysis, enzymatic hydrolysis.*

Lignocellulosic biomass is biomass that is made up principally of lignin, hemicellulose and cellulose. This class of biomass includes woody and fibrous materials from organic sources, agricultural wastes, organic municipal wastes and organic industrial wastes. Depending on origin, the lignin content of lignocellulose varies from about 10 - 25 per cent, the hemicellulose content from about 20 - 35 per cent and the cellulose content from about 35 - 50 per cent (Wyman, 1996).

Lignin is a polymer of phenylpropanoid units interlinked through a variety of nonhydrolysable C - C and C-O-C bonds. It therefore is a complex molecule with no clear chemical definition as its structure varies with plant species. Hemicellulose is an amorphous heterogenous group of branched polysaccharides. Its structure is characterised by a long linear backbone of one repeating sugar type with short branched side chains composed of acetate and sugars. Cellulose is a linear molecule consisting of repeating cellobiose units held together by β - glycosidic linkages (Duff & Murray, 1996). Cellulose is more homogeneous than hemicellulose but is also highly crystalline and highly resistant to depolymerisation. In addition to their rather complex structures, the three components of lignin, hemicellulose and cellulose are tightly bound to each other in the biomass. In fact hemicellulose acts as a bonding agent between cellulose and lignin. In order to convert these biomass to fuel ethanol, the biomass has to be broken up into the individual components first before the molecular chains within each component can be broken up further into simpler molecules. Hemicellulose can be broken up *i.e.* hydrolysed to yield simpler molecules such as arabinose, mannose, glucose, galactose, xylose (the most abundant) and uronic acid, while cellulose can be hydrolysed to yield glucose molecules. Compared to hemicellulose and cellulose, lignin on the other hand cannot be easily hydrolysed. While hemicellulose can easily be hydrolysed with boiling dilute acid, the same cannot be said of cellulose. Currently there exists two broad categories of methods for the hydrolysis of cellulose. One category uses acid (principally H_2SO_4) while a second category uses enzymes called cellulases. Whatever the method employed, the final hydrolysate contains sugars which can then be fermented by micro-organisms to yield ethanol. It should be noted that within each category variations in procedures exist but generally the flow of the various processes are as depicted in *Figure 1*. Also note that depending on the actual procedure adopted, by-products can also be extracted after each of the pretreatment, hydrolysis and fermentation stage. Each of the steps shown in *Figure 1* will be briefly described next.

SIZE REDUCTION

Depending on the type of biomass and the subsequent procedure to be followed, the size of the lignocellulose is usually reduced to a few millimetres to a couple of centimetres in either the wet or dry state.

PRETREATMENT

Again depending on the hydrolysis process to be followed, pretreatment (Duff & Murray, 1996; Hsu, 1996) can be simple or more involved. The purpose of the pretreatment is to render the biomass more amenable to hydrolysis. We describe below some pretreatment processes that have been found to be useful and workable.

(i) Steaming/steam explosion

In this process the biomass is subjected to steaming at a high pressure which is then reduced either slowly or rapidly. The hemicellulose goes into solution while lignin and cellulose remain as solids. This procedure has advanced to pilot scale.

(ii) Hydrothermolysis

In this process, the biomass is cooked in water at high temperatures. The process has not gone much beyond bench scale testing. This process is also known as aqueous fractionation, uncatalysed solvolysis or aquasolv process.

(iii) Dilute acid process

Here the lignocellulose is first treated with a solution of dilute acid. Usually H_2SO_4 is used though other acids such as nitric, hydrochloric, phosphoric and peracetic have also been tried. In this process, the hemicellulose goes into solution while lignin and cellulose remain as solids. For this process corrosion resistant vessels are needed.

(iv) Alkaline process

This is basically a delignification process as the alkali (usually NaOH) used dissolves lignin. The procedure also dissolves hemicellulose, leaving mainly cellulose as a solid.

(v) Organosolv process

Solvents such as acetone, ethanol, methanol and others are used to solubilise the lignin. In the process some hemicellulose are also

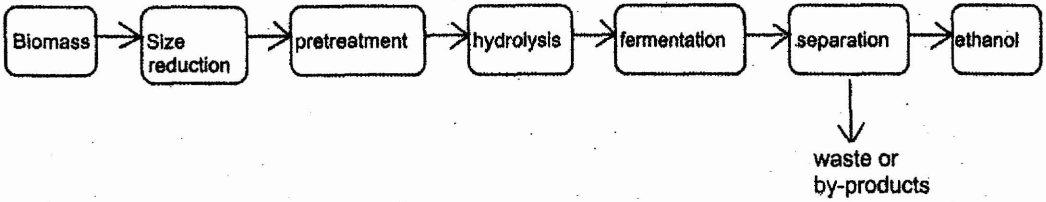

Figure 1 Process flow for converting biomass to ethanol

removed. Since organic solvents are costly and their use requires high pressure equipment, this process is generally perceived as complex and expensive.

(vi) Steam explosion with SO_2 or CO_2

Here, SO_2 or CO_2 is used as catalyst in the steam explosion process. It is found that SO_2 is more effective than CO_2 but the former is highly toxic and therefore must be used with great care. As in steam explosion, hemicellulose goes into solution while lignin and cellulose remain as solids.

HYDROLYSIS

After the various treatments mentioned above, the cellulose in the biomass is now more exposed and should be more amenable to being broken down into simpler sugar molecules. This breaking down process is known as hydrolysis. There are currently two major categories of methods employed. The first and older category of methods uses acid while the second category uses enzymes called cellulases.

(i) Acid hydrolysis

There are several variations to hydrolysing cellulose using acids. However generally when a more concentrated solution of acid is used the temperature required is lower while a more dilute solution of acid will require a higher (usually >200°C) temperature for the hydrolysis. Though different types of acid such as sulphric, hydrochloric, hydrofluoric and nitric acids can be used; because of cost, sulphuric acid is the one most commonly used. While some methods use a single hydrolysing stage, others employ a two or even three stage process. Nowadays most processes incorporate a prehydrolysis or pretreatment step where the relatively easy to hydrolyse hemicellulose is first removed. The different process variations developed have been given names either by the originators or subsequent researchers or users. Some examples are:

Scholler-Holzminden Process
Madison Process
Hokkaido Process
Noguchi - Chisso Process
Udic - Rheinau Process
Scholler - Tornesch Process
Battelle - Geneva Process
Bergius - Rheinau Process
Canada America Sweden Hydrolysis (CASH) Process
Tennessee Valley Authority Process

For a further description of each of the above processes please refer to Schell and Duff (1996), IEA Bioenergy Newsletter (1998), Strickland *et al.* (1988), Saeman and Andreasen, (1954). It should also be mentioned that modifications to some of the above processes have also been initiated by users and researchers.

Compared to enzymatic approaches to be discussed in the next section, acid hydrolysis has a long history. In fact acid hydrolysis of plant lignocellulosic biomass has been known since 1819 (Keller, 1998). During World War I and the worldwide depression of the late 1920's, several plants were built. Those in Germany, Switzerland and Korea used either concentrated hydrochloric acid or the Scholler hot dilute sulphuric acid process. During World War II, full scale plants were built in Mannheim and Regensberg, Germany. These plants operated using the modified Bergius process, but more recently the modified Scholler processes were used in plants in the former Soviet Union, Japan and Brazil (Keller, 1998).

In dilute acid approaches though the

reaction rate is fast the high temperature needed degrades the sugars following their release into solution and as such yields are lower; thus making this approach less attractive. Higher concentration acid approaches provide better yields but the acid used must be recovered for both environmental and economic reasons. For this approach to be viable, the acid recovery system however must be of low cost. Presently several concentrated acid processes are being considered for commercial applications (Wyman, 1996) especially in new pilot plants while older plants tend to employ dilute acid processes.

Listed below are some examples of pilot plants built after the oil crisis of the 1970's.

American Can Company Plug Flow Reactor
Tennessee Valley Authority Pilot Plant
New York University Pilot Plant
Bio-hol/St. Lawrence Reactor
National Renewable Energy Laboratory Pilot Plant

For futher details please refer to Schell and Duff (1996) and the IEA Bioenergy Newsletter (1998). It should be noted that some of the above plants are no longer in operation. Even so a California based company, Arkenol Inc. is still moving ahead with plans to build in Sacramento, California, a commercial scale rice straw-to-ethanol plant using Arkenol's proprietary concentrated acid hydrolysis technology (Arkenol Home Page, 2004). Meanwhile researchers at Universiti Sains Malaysia, have successfully demonstrated that ethanol can be produced with reasonable yields from oil palm wastes *via* sulphuric acid hydrolysis (Yeoh & Lim, 2000). They estimated that if all the lignocellulosic oil palm wastes generated in Malaysia were to be converted to fuel ethanol, the quantity produced would be able to meet about 20 per cent of the total energy requirements of the country's transport sector in 2000. This percentage can be further increased if the xylose fractions are also used.

(ii) Enzymatic hydrolysis

In this process, the breaking up of cellulose into its component sugars is accomplished by the use of enzymes called cellulases which are produced by a variety of bacteria and fungi. Currently the fungus, *Trichoderma reesei* is the most popular microorganism used for cellulase biosynthesis. Other microorganisms studied include *Penicillium*, *Sclerotium rolfii*, *Thermomonospora* and other *Trichoderma* species (Kadam, 1996).

In order for enzymatic hydrolysis to take place satisfactorily, the biomass has to be pretreated with one or more of the pretreatment procedures described earlier. The pretreatments required for enzymatic hydrolysis in general, are more complex and rigorous than those required for acid hydrolysis; otherwise the cellulose will not be accessible enough for the cellulase to act. Pretreatment procedures can also result in the hydrolysis of the hemicellulose component such as in the case of pretreatment with dilute acid at mild temperatures.

Since cellulases have to be produced, enzymatic approaches tend to be more complex than the acid hydrolysis process and also more time consuming as the cellulases need longer time to act. The cellulases are also fairly expensive to produce (Kadam, 1996). However since enzymes are more selective in their action, less degradation products are encountered after the hydrolysis. Another plus point is that there are few or no by-product disposal problems (Kadam, 1996; Himmeh, 1996). The optimum temperature for enzymatic hydrolysis is 45° - 50°C (Philippinis, 1996).

In the last 20 or so years, a lot of effort has been directed at improving the enzymatic hydrolysis process and to date substantial progress has been made. Even so research centres such as the NREL in Colorado, USA, are still active in trying to improve the process further. In view of the progress made, numerous pilot plants have been constructed to demonstrate the feasibility of the enzymatic approach on a commercial scale. Such pilot plants include:

Iotech Pilot Plant
Soustons Pilot Plant in France
The Research Association for Petroleum Alternatives Development (RAPAD) Pilot Plant in Japan.
Voerst-Alpine Pilot Plant
NREL Pilot Plant

Details of these pilot plants can be found in Schell and Duff (1996) and Ballerini *et al.* (1994).

In addition, Iogen Corporation has built a $25 million commercial demonstration plant in Ottawa, Canada, that will convert cellulosic materials to ethanol using an enzymatic process (Kluepfel, 1999). BC International has in 1998 also launched a $90 million industrial - sized plant in Jennings, Louisiana to convert bagasse and rice hulls to ethanol using a genetically engineered organism developed by the University of Florida (Biofuels News, 1998).

It has been estimated that the mid 1990's cost of ethanol production *via* enzymatic - based processes is around 32 US cents per litre at the plant gate thus making it competitive with ethanol from corn for blending with gasoline (Padukone, 1996). To compete with gasoline as a pure fuel, the cost has to be lowered further. Presently the concentrated acid and enzymatic based approaches appear to offer similar projected costs but because of rapid advances in biotechnology, many are of the opinion that enzymatic approaches may have a better future (Wyman, 1996) as evidenced by the commercial ventures mentioned above. Also the US Department of Energy aims to produce ethanol from wood at a cost of 20 US cents per litre by 2005 and 14 US cents per litre by 2030. The production targets, using enzymatic hydrolysis, are about 9 billion litres of ethanol by 2005 and as much as 85 billion litres by 2030 (Chaturvedi, 2004).

FERMENTATION

After hydrolysing the cellulose, micro-organisms are then used to ferment the glucose molecules into ethanol. This process is a well proven and established process and baker's yeast (*Saccharomyces cerevisiae*) is the organism most commonly used (Philippinis, 1996; Picataggio & Zhang, 1996). Another organism that is gaining recognition is *Zymomonas mobilis* (Picataggio & Zhang, 1996). The optimum temperature for the fermentation process is around 30°C (Philippinis, 1996). In order to increase ethanol yield and therefore make the biomass-to-ethanol route more economically viable, the sugar molecules, principally xylose, from the hemicellulose component of the biomass, should also be fermented to produce ethanol. It should be noted that whether the acid or enzymatic approach is followed, xylose is already in solution.

Several microorganisms are known to be able to ferment the xylose with *Pichia stipitis* being a popular choice at this moment in time (Picataggio & Zhang, 1996; McMillan, 1996). With *P. stipitis*, aeration is required. The recombinant bacteria *Escherichia coli* and *Klebsiella oxytoca* and strains of *S. cerevisiae* and *Z. mobilis* are also gaining recognition (Picataggio & Zhang, 1996; Dumsday *et al.*, 1997). With recombinant enteric bacteria, high yield and productivity can be achieved under anaerobic conditions (McMillan, 1996). For more effective conversion, the hydrolysates should preferably be detoxified. For this a variety of methods can be used; they include overliming and heating, steam stripping, rotoevaporation, ion exchange, extraction and treatment with activated carbon and molecular sieves (McMillan, 1996). In addition research is also being conducted to produce microorganisms that are able to ferment both xylose and glucose simultaneously and efficiently (Picataggio & Zhang, 1996). Presently genetically engineered *E. coli*, *K. oxytoca* and *Z. mobilis* strains appear to be promising candidates (Padukone, 1996).

SEPARATION

After the fermentation process, the ethanol produced has to be separated from the fermentation broth. Various procedures such as distillation, use of membranes and absorbents *etc.* can be used but by far the distillation procedure is the most popular as this process has been well proven to work.

OTHER TECHNOLOGIES AND PROCESSES

Even though the schematic diagram of *Figure 1* is the process flow generally adopted for the conversion of lignocellulosic biomass to ethanol, it must be emphasised that currently other methods and procedures are still being actively investigated. The aim is to try to increase the ethanol yield, while at the same time reduce material inputs and process time and hence the overall cost of production. The better known examples of such methods include (i) simultaneous saccharification and fermentation (SSF) and (ii) direct microbial conversion (DMC).

(i) Simultaneous saccharification and fermentation (SSF)

As the name implies, SSF technology combines the enzymatic hydrolysis of cellulose and the glucose fermentation steps into one step. The temperature for optimal operation is 37° - 38°C (Philippinis, 1996). SSF technology is generally viewed as a promising way to convert cellulose into ethanol because of its reduced number of steps (Philippinis, 1996). The technology however is still actively being researched and developed. Presently pilot scale systems are available to demonstrate the feasibility of this technology. An example is the Gulf/University of Arkansas Pilot Plant. Another pilot plant, the Katzen Pilot Plant constructed by Ralph Katzen Associates International together with the University of Arkansas and Procter & Gamble also operates using the SSF technology (Schell & Duff, 1996).

(ii) Direct microbial conversion (DMC)

This process uses cellulase - synthesising bacteria such as *C. thermocellum*, *C. thermosaccharolyticum* and *Thermoanaerobacter ethanolicus* and fungi such as *Fusarium oxysporum* to directly convert cellulose to ethanol. The technology therefore combines all three processes (cellulase production, cellulose hydrolysis and fermentation) into one step (Padukone, 1996). The problems encountered in this technology include low ethanol yield and high by-product formation (Philippinis, 1996). Thus the potential of this technology may be limited.

GENERAL DISCUSSION AND CONCLUDING REMARKS

As mentioned in the Introduction, the process flow shown in *Figure 1* is the general procedure adopted. However depending on the nature and composition of the biomass, certain processes such as size reduction, may be omitted and more than one of the processes shown in *Figure 1* may be combined. By-products can also be separated or extracted after each process.

Since large quantities of biowastes, such as those from the oil palm industry, are produced in our country, a large scale systemic study on the feasibility of converting these wastes to fuel ethanol ought to be carried out, though, as mentioned earlier, preliminary studies have been initiated at Universiti Sains Malaysia. Such a large scale study would ultimately indicate which of the two hydrolysis routes is more appropriate for each type of biowastes the country generates.

With the rapid progress in biotechnology and process engineering the above account, though brief, does indicate that producing fuel ethanol from lignocellulosic biomass is fast becoming an economically viable option and it may be wise for our country to take cognisance of this.

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