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Introduction

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Welcome to “Aqueous Pretreatment of Plant Biomass for Biological and Chemical Conversion to Fuels and Chemicals.” This book provides insights into thermochemical preparation of cellulosic biomass such as wood, grass, and agricultural and forestry residues for aqueous conversion to fuels and chemicals as well as economic and analysis information that is broadly applicable to a wide range of aqueous biomass operations. Historically, acid catalyzed hydrolysis of biomass goes back to the early nineteenth century [1], when the emphasis was on aqueous-processing of biomass in concentrated acid or dilute acid at higher temperature to break down cellulose into glucose that could be fermented into ethanol for use as a fuel [2,3]. Because most of the hemicellulose sugars are destroyed at dilute acid conditions that realize high glucose yields from cellulose, pretreatment with dilute acid at milder conditions was employed to maximize yields of hemicellulose sugars (provided they were removed prior to treating the cellulose [4]). Then, most of the cellulose was left in the solids and could be broken down with dilute acid at more harsh conditions to fermentable glucose without sacrificing much of the hemicellulose sugars [5]. A similar approach was applied commercially to break down hemicellulose in corn cobs, sugar cane bagasse, and other hemicellulose-rich types of cellulosic biomass into xylose and arabinose sugars, and react these sugars further to marketable furfural [6]. In this case, the cellulose, lignin, and other components left in the solids were usually burned for heat and power. Application of milder conditions for hemicellulose breakdown was later found to be effective in opening up the biomass structure so enzymes could achieve high glucose yields from the recalcitrant cellulose left in the solids [5,7,8]. More recently, hemicellulose conversion to sugars or furfural has been employed followed by heterogeneous catalysis to produce hydrocarbons from biomass that are compatible with existing fossil-resource-based fuels and chemicals [9–11]. In this case, even harsher dilute acid conditions than applied to release glucose from cellulose could then be applied to the remaining cellulose-rich solids to generate 5-hydroxymethyl furfural and levulinic acid, desirable

precursors for catalytic conversion into hydrocarbon fuels and chemicals. In a sense, technology for thermochemical breakdown of cellulosic biomass with dilute acid has come full circle from its beginnings, albeit to serve different downstream processes.

The operation to prepare biomass for downstream aqueous biological or catalytic processing is typically called pretreatment and is critical to achieving high product yields that can foster the emergence of biofuels and biochemicals industries based on biological or catalytic conversion of plants. However, the range of technologies has become broader than just the reaction of hemicellulose in dilute acid and now includes operations that also focus on lignin removal [12–14]. For biologically based processes, disruption of hemicellulose or lignin (and not removal) may also be adequate to realize high sugar yields from biomass in enzymatic operations. Furthermore, a wide range of combinations of reaction temperatures, pH values, and times can be effective in preparing biomass for downstream processing, depending on the technologies being applied [15–17]. Some of these aqueous pretreatments build from analogous industrial operations such as removal of lignin by reaction of biomass with caustic for the pulp and paper industry. We can therefore now define aqueous pretreatment as the reaction of cellulosic biomass at conditions that result in the highest possible yields in subsequent biological, catalytic, or thermochemical processing.

The goal of this introductory chapter is to summarize some of the key aspects of cellulosic biomass and its aqueous pretreatment to make it compatible with downstream biological, catalytic, or thermochemical processing to provide an historical perspective for the chapters in this book and its organization. This chapter will start by providing a sense of what we mean by cellulosic biomass and why it is a vital resource for sustainable production of organic fuels and chemicals. This overview will be followed by a summary of key biomass features, including its composition. An overview will then be given of how biomass lends itself to biological and catalytic aqueous processing and the important challenges hindering commercial applications. Against this background, criteria for successful pretreatment will be outlined. An overview of various pretreatment technologies will then provide a sense of options that have been investigated over the years and the rationale behind the emphasis on thermochemical pretreatments in this book. In addition, other aspects that can influence pretreatment effectiveness will be mentioned, along with limitations in our experience with pretreatment. The chapter will end with an outline of the chapters that follow to help the reader utilize the information in the book.

1.1 Cellulosic Biomass: What and Why?

The word biomass encompasses any biological material derived from living or recently living organisms. The term could therefore apply to both animal and vegetable matter. However, this book focuses on cellulosic biomass, the structural portion of plants, as a resource for the production of fuels and chemicals. Plant/cellulosic biomass contains carbon, hydrogen, and oxygen, plus typically much lower amounts of nitrogen, phosphorous, minerals, and other ingredients. The sun's energy drives the formation of plant biomass while releasing oxygen through the photosynthetic reaction of water with carbon dioxide. The late Dr Ray Katzen, a giant in the field of industrial biomass conversion, termed cellulosic biomass as C-water – CH_2O – in reference to the building block from which biomass sugars are made. If biomass or materials derived from biomass are burned, oxygen in the air combines with the carbon and hydrogen in biomass to release carbon dioxide and water, reversing the reactions through which plant matter was formed originally. However, as long as new biomass is planted to replace that burned or otherwise utilized, this carbon cycle results in no net change in the amount of carbon dioxide in the atmosphere. This feature of using biomass distinctly contrasts with burning fossil fuels, in which carbon from below the ground continually accumulates in the atmosphere. The powerful natural carbon recycle provides the potential for fuels production from cellulosic biomass to avoid contributing to the net accumulation of carbon dioxide in the atmosphere, a major driver of global climate change [18–21].

Biomass can fill a unique niche for sustainably meeting human needs. The sustainable resources are sunlight, wind, ocean/hydro, geothermal, and nuclear, and societal needs can be grouped as food, motor-driven devices, light, heat, transportation, and chemicals [22]. Electricity and thermal energy can be made from all sustainable resources as primary intermediates for human needs but only sunlight can support growth of biomass, the other primary intermediate. Biomass alone among sustainable resources can be transformed into feed for animals, human food, and organic fuels, chemicals, and materials. Plant materials could have a much greater impact if vast, low-cost untapped sources of cellulosic biomass such as agricultural and forestry residues, portions of municipal waste, and dedicated crops could be inexpensively converted into a range of fuels and commodity chemicals in large-scale biorefineries [23]. In fact, inexpensive transformation of biomass into liquid fuels and commodity chemicals will be essential if society is to sustainably and economically meet such needs [24–26].

Although the term cellulosic biomass may not be a household word, it represents the structural portion of a large group of well-known plants. Common examples include agricultural wastes such as corn stalks and corn cobs (the two together being termed corn stover) and sugar cane bagasse that are left after removal of targeted food and feed products. Forestry residues represent another familiar example of cellulosic materials as represented by sawdust, bark, and branches left after harvesting trees for commercial operations such as making paper and wood products. Large portions of municipal solid wastes, including waste paper and yard waste, are also cellulosic biomass. Paper sludge results from fines from plant biomass not captured in the final product [27,28]. Although such existing cellulosic resources can cumulatively represent a substantial resource that could provide an effective platform from which to launch a biomass-based industry, energy crops will be ultimately needed to meet the huge demand for organic fuels and chemicals. In this vein, various types of grasses can prove to be valuable feedstocks with fast-growing herbaceous plants such as switchgrass and *Miscanthus* being prominent examples. In addition, various trees such as poplar and eucalyptus have the high productivities desirable to maximize production potential from limited available land. Taken together, it has been estimated that the future availability of biomass for energy production in the United States could be on the order of 1.4 billion dry tons of biomass, enough to displace over 100 billion gallons of gasoline of the approximately 140 billion gallons now used in the United States [29,30]. Biomass-based fuels could make an even bigger impact if the country were to substantially reduce fuel consumption by driving more efficient vehicles and use more public transportation.

In addition to being widely available, having the potential to reduce greenhouse gas emissions, and being uniquely suited to sustainable production of liquid fuels, cellulosic biomass is inexpensive. For example, cellulosic biomass costing \$60 per dry ton has about the same cost per unit mass as petroleum at about \$7 per barrel. Of even more relevance for fuels production, this biomass price would be equivalent to petroleum at about \$20/barrel on the basis of equivalent energy content [27,31]. The resource itself is therefore low in cost, and the challenge is how to inexpensively transform cellulosic biomass into fuels.

1.2 Aqueous Processing of Cellulosic Biomass into Organic Fuels and Chemicals

A variety of pathways can be applied to convert cellulosic biomass into fuels and chemicals [11]. For example, cellulosic biomass can be gasified to generate carbon monoxide and hydrogen. This mixture, called syngas, can in turn be catalytically converted into diesel fuel, methanol, or other products. Pyrolysis by heating biomass in the absence of air can generate oils that must be upgraded to have suitable fuel properties and be more compatible with conventional fuels. Biomass could be liquefied by application of heat and hydrogen under pressure. For such thermal routes, a proximate analysis of biomass composition may be useful to support design of a process. For example, a typical proximate analysis of switchgrass could be about 13.7% fixed carbon, 73% volatile matter, 4.9% ash, and 8.4% moisture [32]. The higher heating value could be about 17.9 MJ/kg. However, the elemental composition of biomass is likely to be more

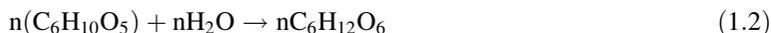
informative in that it allows development of more in-depth material and energy balances. In this case, a representative elemental analysis of switchgrass could include about 46.8% carbon, 5.1% hydrogen, 42.1% oxygen, less than 0.6% nitrogen, about 0.1% sulfur, and 5.3% minerals/ash, all being on a mass basis [32].

In reality, cellulosic biomass is more complex than simple proximate or elemental analyses suggest, with their structures evolved to support key plant functions [33]. Although the wide range of plant materials represented by cellulosic biomass are distinct in physical appearance, they all share similar structural make-ups. Generally, the most abundant portion is cellulose; about 35–50% of the weight of many plants comprises cellulose. Cellulose is a polymer of glucose sugar molecules linked together in long, straight parallel chains that are hydrogen-bonded to one another in a crystalline structure to form long fibers. Another roughly 12–25% of cellulosic biomass is a sugar polymer known as hemicellulose, which can consist of the five sugars arabinose, galactose, glucose, mannose, and xylose along with various other components such as acetyl groups and pectins [34,35]. The proportion of these components in hemicellulose varies among plants and, unlike cellulose, hemicellulose is branched and not crystalline. The other significant fraction of cellulosic biomass is lignin, a complex phenyl propene compound that is not made of sugars and whose chemical composition varies with plant type [33,36]. Cellulosic biomass also contains lesser amounts of other compounds that may include minerals/ash, soluble sugars, starch, proteins, and oils. Although often overlooked in the discussion of biomass conversion, these components are also vital to plant functions.

Aqueous processing targets processing of cellulosic biomass in water to convert the structural components in biomass into compounds dissolved in water which we call reactive intermediates (RIs) that, in turn, can be biologically, catalytically, or thermochemically converted into fuels or chemicals. Thus, biomass is broken into the basic building blocks from which it is made and not all the way down to simple molecules. For example, the arabinose and xylose in hemicellulose are five carbon sugar isomers that can be linked together in a chain n units long to form $n(\text{C}_5\text{H}_8\text{O}_4)$. As noted above, acids or enzymes can catalyze the breakdown of such chains in water to release the individual five carbon sugars from which they are made by the following hydrolysis reaction:



Similarly, acids or enzymes can catalyze hydrolysis of the six carbon sugars that comprise a portion of hemicellulose and all of cellulose (glucose) into the sugar isomers glucose, galactose, or mannose as follows:



The arabinose and xylose released from reaction (1.1) and galactose, glucose, and mannose released by reaction (1.2) can all be fermented to ethanol or other products through a choice of suitable organisms. For example, industrial yeast strains such as *Saccharomyces cerevisiae* or other yeast naturally ferment glucose and the other six carbon sugars into ethanol. Furthermore, although native yeast cannot ferment the five carbon sugars arabinose and xylose to ethanol with high yields, various bacteria such as *Escherichia coli* and yeast including *Saccharomyces cerevisiae* have been genetically engineered so they now produce ethanol from these sugars with high yields [37–40]. We can therefore view these sugars as reactive intermediates that can be biologically converted into ethanol and other final products.

A variety of acids including sulfuric, nitric, and hydrochloric have been applied to hydrolyze hemicellulose to its component sugars with yields of about 80–90% of theoretical or more, feasible in simple batch or co-current flow operations [41,42]. Dilute acids can also hydrolyze cellulose to glucose, but glucose yields are limited to about 50% of theoretical for practical operating conditions [2,43]. Enzyme

catalyzed breakdown (hydrolysis) of cellulose to glucose has therefore emerged as a leading option for making commodity products because nearly theoretical glucose yields vital to economic success are possible [22,27]. Furthermore, enzyme-based processing costs have been reduced by about a factor of four [44–51], and many of the additional advances needed to make the technology competitive are achievable through application of the powerful new and evolving tools of biotechnology [48,49,52–54]. Another benefit of high-selectivity biological conversion and particularly enzymatic catalysis is minimal waste generation, reducing disposal problems. Although efforts have focused on ethanol production, a range of fuels, chemicals, and materials can be biologically derived from the same sugar intermediates [24,25,55]. However, the key obstacle to commercial use of enzymes for release of sugars from cellulosic biomass is the high doses and resulting high costs for cellulase and hemicellulase [27,31,55,56]. The most critical need to achieve low production costs is therefore the reduction of biomass recalcitrance as the major obstacle to low sugar costs [27,56].

Although sugars can be fermented into a wide range of compounds that are valuable fuels and chemicals, many are oxygenated and differ from currently employed hydrocarbons. For example, ethanol is a high octane fuel with many superior properties to gasoline, with the result that it is the fuel of choice for the Indianapolis 500 and other races for which speed and power are vital. Ethanol is also much less toxic than gasoline as evidenced by the fact we drink beer, wine, mixed drinks, and other beverages containing ethanol while no beverages contain gasoline. The fact that ethanol is different from gasoline concerns many users, however. For example, ethanol has a somewhat lower energy density, tends to separate into water when water is present, and has different solvent properties from gasoline. Thus, many desire hydrocarbon fuels that are completely fungible with the current petroleum-based infrastructure. This preference for hydrocarbons is appropriate for aviation, for example jet fuel which needs the highest possible energy density. Similarly, hydrocarbons have important advantages in compression ignition engines that are important in powering large trucks, earth-moving equipment, and other heavy-duty vehicles.

Whatever the rationale, aqueous biomass streams are now being processed into RIs including furfural, 5-hydroxymethylfurfural (5-HMF), and levulinic acid for catalytic conversion into hydrocarbon “drop-in” fuels by novel processes [9,10]. Aqueous catalysis can build off many of the same pretreatment technologies developed for biological conversions, but without enzymes or fermentations. To support catalytic processing, enzymes or acid catalyze hydrolysis of the cellulose and hemicellulose into their sugar monomers in the same way as for biological conversion. However, dilute acids also catalyze dehydration of the sugars into sugar alcohols that can be aldol condensated and hydrogenated into RIs and light alkanes by homogeneous/heterogeneous catalysts [9]. The catalysts used for these reactions include acids, bases, metals, metal oxides [10,57,58], and multifunctional catalysts. For example, ruthenium/carbon (Ru/C) and platinum/zirconium phosphate (Pt/ZrP) catalysts hydrodeoxygenate aqueous streams of xylose to xylitol at 393 K and xylitol to gasoline range products at 518 K. Bimetallic PtSn catalysts selectively hydrogenate furfural to furfural alcohol, which acids can further hydrolyze to levulinic acid (LA), a reactive building block for hydrocarbon fuels. LA can in turn be converted into gamma-valerolactone (GVL) over Ru/C catalyst. Further, GVL can be converted to equimolar amounts of butene and carbon dioxide gases through decarboxylation at elevated pressures over a silica/alumina catalyst. This stream can in turn be converted into condensable alkenes by the application of an acid catalyst (e.g., H ZSM-5, Amberlyst-70) that links butene monomers to achieve molecular weights that can be compatible with gasoline and/or jet fuel applications [59].

1.3 Attributes for Successful Pretreatment

From the above discussion, aqueous pretreatment can be applied to prepare cellulosic biomass for subsequent enzyme or acid catalyzed reactions to release sugars for fermentation to ethanol or other products.

In such cases, the primary goal for pretreatment is to work with downstream operations to achieve the highest possible product yields at the lowest costs; a variety of pretreatment approaches are promising [15–17,60]. Aqueous pretreatment is also applicable in preparing cellulosic biomass for catalytic reaction, with the goal again being to achieve the highest possible product yields and lowest costs. However, current pretreatment approaches favored for catalytic processing employ dilute acid to remove hemicellulose with high sugar or furfural yields. In addition, dilute acid can also be employed for subsequent reaction of the cellulose-enriched solids from pretreatment into HMF and/or levulinic acid. Aqueous pretreatment of biomass to support catalytic conversion can therefore avoid the high costs of enzymes that have hindered commercialization of biological routes to fuels and chemicals.

Against this background, several key attributes are vital for pretreatment to be promising for application to biological or catalytic conversion of cellulosic biomass to fuels and chemicals. Because milling of biomass to small particle sizes is energy intensive and introduces extra equipment costs [61,62], pretreatment technologies that require limited size reduction are desirable. In the case of enzymatic conversion, pretreatment must open up the biomass structure to make cellulose accessible to enzymes so they can achieve high yields from the pretreated solids and recover sugars released in pretreatment with high yields. To support catalytic processing, pretreatment must achieve high sugar or furfural yields from hemicellulose as well as serve subsequent reactions to target RIs. Regardless of the downstream operation, the concentration of RIs should be as high as possible to ensure that product concentrations are adequate to keep recovery, process equipment, and other downstream costs manageable. The requirements for chemicals in pretreatment and subsequent neutralization and conditioning for downstream operations should be minimal and inexpensive, or the chemicals should be easily recovered for reuse. Pretreatment reactors should be low in cost through minimizing their volume, requiring low pressures and temperatures, and avoiding the need for exotic materials of construction due to highly corrosive chemical environments. In addition, the pretreatment chosen must work cooperatively with other operations. For example, a pretreatment operation that separates hemicellulose sugars from glucose from cellulose may be preferred to avoid preferential glucose fermentation and associated lower yields from hemicellulose sugars due to diauxic effects. The liquid stream from pretreatment must be compatible with subsequent steps following a low-cost high-yield conditioning step. In fact, it is highly desirable to employ pretreatments that produce streams that require no conditioning to reduce costs and reduce yield losses. Any chemicals formed during hydrolyzate conditioning in preparation for subsequent steps should not present processing or disposal challenges (e.g., gypsum formed by neutralization of sulfuric acid with calcium hydroxide). An innovative pretreatment could recover lignin, protein, minerals, oils, and other materials found in biomass for use as boiler fuel, food, feed, fertilizers, and other products in a biorefinery concept that enhances revenues [63]. Such synergies would leverage biomass impact and reduce land requirements, enhancing sustainability [24,25,27,55]. Consequently, attention must be given to advancing pretreatment to make aqueous processing of biomass competitive for large-scale sustainable applications in an open market [64,65]. A number of reviews of pretreatment, enzymatic hydrolysis, and catalytic processing provide historic perspectives [e.g. 12,13,31].

In choosing a pretreatment technology, high product yields must be met to distribute total costs over as much product as possible. In addition, the capital and operating costs for pretreatment must be kept low without sacrificing product yields. We could therefore say that the best pretreatment would be free and have no costs or unwanted impacts on other operations; unfortunately however, pretreatment has been projected to be the most expensive single operation in overall biological processing in some studies [66]. Because yields suffer without pretreatment, other studies have shown that overall product unit costs are higher without pretreatment than with it, leading this author to state that “the only operation more expensive than pretreatment is no pretreatment” [31]. Ultimately, the choice of pretreatment is governed by costs of the overall process and not just the pretreatment operation [67–71].

1.4 Pretreatment Options

Over the years, a number of aqueous-based pretreatment technologies have been investigated in the search for a low-cost approach that can realize high yields of final products from both the cellulose and hemicellulose fractions [72]. Most of these have focused on supporting subsequent enzymatic hydrolysis, with only limited recent work supporting catalytic processing. Reviews have classified these pretreatment methods as (1) physical, (2) biological, and (3) chemical.

Physical pretreatments include size reduction by devices such as hammer mills, knife mills, extruders, disc refiners, and planers. Mechanical decrystallization by ball, roll, dry, and colloid mills are physical pretreatments that can increase enzymatic hydrolysis yields. Thermal pretreatment by freeze/thaw, pyrolysis, and cryomilling are also classified as physical pretreatments, as are radiation with gamma rays, microwaves, electron beams, and lasers. Many physical pretreatments are not sufficiently effective in achieving high yields, and their operating and/or capital costs are often high [73–79]. Overall, such methods are not yet considered practical to support biological processing and do not produce the RIs needed for catalytic methods. These methods are therefore not covered in depth in this book, but other sources can be checked for more information for those wishing to explore these technologies further [80].

Biological pretreatment of biomass offers some conceptually important advantages such as low chemical and energy use. Generally, organisms are sought that will preferentially attack lignin to open up biomass for subsequent attack by enzymes. Various fungi including *Fomes fomentarius*, *Phellinus igniarius*, *Ganoderma applanatum*, *Armillaria mellea*, and *Pleurotus ostreatus* are typical choices. Unfortunately, to date, biological methods tend to suffer from poor selectivity in that organisms consume cellulose and hemicellulose, hurting product yields. In addition, they require long times and are hard to control. Overall, because no biological system has been demonstrated to be effective [81–85], they are not considered further in this book and the reader should consult other sources for additional insights [86–89].

Chemical pretreatments make up the third and final class of options that employ a range of different chemicals to prepare biomass for subsequent operations [12,13]. Most also include raising the temperature to the range of 140–210 °C or so and are labeled as thermochemical pretreatments. The result is a broad range of chemical concentrations, temperatures, and times that have been applied for biomass pretreatments. Oxidizing agents such as peracetic acid, ozone, hydrogen peroxide, chlorine, sodium hypochlorite, and chlorine dioxide as well as oxygen and air have been employed for thermochemical pretreatment. Another set of options revolves around concentrated acids including sulfuric (55–75%), phosphoric (79–86%), nitric (60–88%), hydrochloric (37–42%), and perchloric (59–61%). Several solvents are effective in dissolving cellulose to improve its accessibility to enzymes, with examples being the inorganic salts lithium chloride, stannic chloride, and calcium bromide, as well as such amine salts as cadmium chloride plus ethylenediamine (cadoxen) and cobalt hydroxide plus ethylenediamine (cooxen). Biomass can also be delignified and fractionated in organosolv pretreatments that employ methanol, ethanol, butanol, or triethylene glycol. Cellulose modification to carboxymethyl cellulose, viscose, or mercerized cellulose provides another thermochemical pretreatment path. The addition of alkaline compounds such as sodium hydroxide, potassium hydroxide, calcium hydroxide, and amines has been employed to open up cellulosic biomass by removing a large portion of lignin. Kraft and soda pulping provide established routes to pretreat biomass at these higher pH levels. Ammonia provides a versatile pretreatment chemical in that it can be applied at gaseous, liquid, aqueous, or supercritical conditions at various moisture levels. Dilute sulfuric or nitric acids do a good job of removing hemicelluloses, as do gaseous hydrochloric acid and sulfur dioxide. In addition, gaseous nitrogen dioxide and carbon dioxide have been tested to reduce the pretreatment pH, although yields are not nearly as high as possible with stronger acids. Perhaps the simplest pretreatment option is to heat biomass with steam or just hot water to break down hemicellulose and dislodge lignin. This approach is sometimes classified as a physical method in that only heat is applied, but it has also been grouped with

thermochemical pretreatments in light of the belief that acetic and other acids released from hemicellulose during pretreatment help catalyze hydrolysis to sugars in what is termed as autohydrolysis. Unfortunately, autohydrolysis does not achieve as high hemicellulose sugar yields as possible with stronger acids.

A number of pretreatment leaders formed a Biomass Refining Consortium for Applied Fundamentals and Innovation (CAFI) in 2000 and worked as a team for over a decade to compare results from the application of leading pretreatment technologies to biological conversion on a consistent basis. The pretreatments studied were based on dilute sulfuric acid, sulfur dioxide, neutral pH, liquid ammonia, ammonia fiber expansion (AFEX), and lime [15,17,68]. The first project focused on application of these pretreatments to corn stover through support from the US Department of Agriculture Initiative for Future Agricultural and Food Systems (IFAFS) Program, and the Office of the Biomass Program of the US Department of Energy supported two subsequent projects on pretreatment of poplar wood and switchgrass. A surprising finding of these three studies was the similarity in results between thermochemical pretreatments spanning a wide pH range from low values with dilute sulfuric acid or sulfur dioxide to high pH values with lime. Yields were particularly similar and high with corn stover for all pretreatments and nearly the same high values for switchgrass across the entire pH range. Total sugar yields from pretreatment together with enzymatic hydrolysis were more variable with poplar wood but even then were similarly high for lime and sulfur dioxide, the extremes in pH. The CAFI studies pointed out that pretreatment effectiveness could not simply be related to process conditions, but that substrate–pretreatment–enzyme interactions are complex. Thus, more detailed research is still needed to better understand how to open up the biomass structure to achieve high yields from the combined operations of pretreatment and enzymatic hydrolysis.

1.5 Possible Blind Spots in the Historic Pretreatment Paradigm

Some very important points should be kept in mind when judging and selecting pretreatment technologies. First, almost all of the past development efforts focused on pretreatment prior to enzymatic hydrolysis, with far less effort devoted to pretreating biomass for catalytic conversion. Thus, consideration of different pretreatment perspectives could be beneficial for the latter. A second vital point is that most of the pretreatment work for biological conversion has evaluated pretreatment effectiveness in terms of yields of sugars by subsequent application of fungal enzymes to the pretreated solids. Furthermore, a large portion of the evaluations of the effectiveness of pretreatment in terms of subsequent enzymatic hydrolysis have been based on high enzyme loadings that would be commercially impractical. Far more work is needed to understand how pretreatments perform at lower enzyme loadings and what features of the pretreated substrate limit high yields. In addition, very little attention has been given to determining relationships among substrate types and features, pretreatment types and conditions, and performance with other biological systems. For example, some bacteria such as the thermophile *Clostridium thermo-cellum* produce a complex cellulosome enzyme structure that may be more effective in hydrolyzing hemicellulose and cellulose into their component sugars with the same organism also fermenting the sugars released to final products. This simultaneous enzyme production and fermentation feature has been called consolidated bioprocessing or CBP. The close association of the enzyme-producing CBP organism with the cellulosome has also been shown to offer significant advantages [90–92]. Another important point concerns the feedstocks pretreated. Although a range of hardwoods, grasses, softwoods, forestry and agricultural residues, and municipal solid wastes have been subjected to pretreatment followed by enzymatic hydrolysis, much less effort has been devoted to determining if particular substrate features would enhance pretreatment performance. Overall, little is known about possible synergies among feedstock features, pretreatment types and conditions, and microbial systems that would greatly enhance yields while simplifying (or possibly eliminating) pretreatment and reducing enzyme loadings, therefore significantly cutting costs.

1.6 Other Distinguishing Features of Pretreatment Technologies

Pretreatment technologies can also be differentiated in ways other than whether they are biological, chemical, or physical or the type of additive used. For example, almost all laboratory experiments are conducted under batch conditions in which all contents are loaded into a reactor at the beginning where they are heated up to some target temperature, held at that temperature for a set period of time, cooled back to room temperature, and then removed for analysis and evaluation. On the other hand, many commercial ventures prefer continuous operations to obtain higher productivities by avoiding heat-up and cool-down times and non-productive periods between batches for emptying and filling reactors, as well as better heat integration. Accordingly, continuous pretreatments are often used with co-current flow of the solids and liquid; the results can be quite similar to those for batch operations if the solids and liquid move as a plug. However, high solids concentrations are also preferred to provide higher sugar concentrations from pretreatment and enzymatic hydrolysis and reduce thermal loads, and cellulosic biomass has little free liquid at such conditions [93–95]. Moving solids of this consistency presents significant challenges, particularly at high temperatures and pressures, and residence times are likely to be variable. Thus, continuous pretreatment performance may be poorer than would be expected from results with laboratory batch systems, and new tools are needed to accurately predict commercial performance.

A number of other operational features can influence performance. For example, some laboratory research has shown that flow of water through a fixed bed of biomass can remove more lignin and hemicellulose and achieve better yields from pretreatment and enzymatic hydrolysis than possible in a batch system operated at similar temperatures and times [96–99]. However, most data from such flowthrough systems has been derived from the use of finely ground biomass, and it is not known how well such systems will perform with larger-sized particles that are more commercially relevant. Bench- and pilot-scale countercurrent pretreatment systems have also shown performance advantages compared to batch operations [100], but moving solids and liquids in opposite directions at high temperatures and pressures at a large commercial scale presents challenges. Methods applied to heat up and cool down biomass can also be very influential, in that variations in temperature histories with time and space can markedly change performance. Washing pretreated biomass with hot water could also improve performance.

1.7 Book Approach

The above information presents an idea of the lay of the land for this book, and has hopefully piqued your appetite for learning more about these and other topics relevant to pretreatment. As noted at the start of this chapter, the aim of the book is to provide comprehensive information that can support research, development, and application of aqueous pretreatment technologies. Experts on biomass pretreatment, conversion, and analysis were invited to author the following 22 chapters to cover the wide range of topics appropriate to the field. These lead authors were responsible for the content of each chapter and in many cases enlisted co-authors. Their intent was to provide solid platforms from which others could understand the importance of pretreatment, developments in the field, fundamentals of the technologies, key attributes and limitations, opportunities for advances, analysis methods, and needs for additional research and development (R&D). Authors were therefore urged to focus on such things as integration into the overall process, reaction kinetics, reaction stoichiometries, reaction conditions, effects on key biomass components, component removal vs. times and temperatures, and equilibrium considerations as appropriate to the chapter topic. This could also include considerations for integration with key upstream and/or downstream operations and their interactions, such as pretreatment with enzymatic hydrolysis. It was also intended that each chapter provides a perspective on the entire topic and facts and not focus on developments in one laboratory or promote

particular technologies, allowing the reader to draw their own conclusions. A particularly important goal was to provide comprehensive references to support key points and allow the reader to obtain additional insights beyond those possible in a chapter of limited length.

1.8 Overview of Book Chapters

As shown in the Table of Contents, this book provides chapters to help the reader understand the unique role of the biomass resource in sustainable fuels production, its composition and structure relevant to pretreatment, the context of aqueous biological and catalytic processing of biomass, features of prominent thermochemical pretreatment technologies, comparative data on application of leading pretreatments to a range of biomass types, economic factors to be considered in pretreatment selection, analytical methods for measuring biomass composition, and experimental systems for pretreatment and enzymatic hydrolysis.

Chapter 2 provides insights into the importance and uniqueness of cellulosic biomass as a resource to support sustainable production of organic fuels and chemicals. Chapter 3 then provides a perspective on the composition of biomass and resulting challenges its recalcitrance presents to conversion. Chapter 4 focuses on biological conversion of cellulosic biomass, with emphasis on challenges facing its incorporation with enzymes and fermentative organisms. An overview of aqueous phase catalytic processing of streams from pretreatment of cellulosic biomass, providing a perspective on the needs for this emerging application, is presented in Chapter 5. Next, fundamental insights are provided on low pH pretreatment and how it can serve both biological and catalytic processing to fuels and chemicals as well as applied to release glucose, 5-HMF, and levulinic acid from cellulose in Chapter 6. Chapters 7 and 8 provide insights into pretreatment fundamentals at nearly neutral pH and high pH to support biological conversion. Chapters are also devoted to outlining fundamental features for pretreatments by AFEX (Chapter 9), biomass fractionation (Chapter 10), and ionic liquids (Chapter 11). Armed with this background, in Chapter 12 the reader is given a summary of data developed for application of leading thermochemical pretreatment technologies to corn stover, poplar wood, and switchgrass, with Chapter 13 providing insights into how enzyme formulations must be tailored to pretreatment type to realize high yields. Chapter 14 provides fundamental insights into how physical and chemical features of pretreated biomass impact sugar release. Cost comparisons for integration of leading pretreatment technologies into biological conversion processes are offered in Chapter 15, and opportunities are defined to reduce conversion costs. Chapters 16, 17, 18 and 19 describe analytical methods that can track changes in biomass composition and other features in pretreatment and enzymatic hydrolysis. Finally, Chapters 20, 21, 22 and 23 are devoted to describing experimental systems that are applicable to pretreatment and enzymatic hydrolysis of biomass, covering scales from multiwell plates to pilot plant operations.

We sincerely hope that the reader finds this book a useful tool to better understand pretreatment of cellulosic biomass, including its importance and insights into leading thermochemical technologies as well as analytical and other supporting methods applicable to any pretreatment of cellulosic biomass.

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