

Strengths, challenges, and opportunities for hydrothermal pretreatment in lignocellulosic biorefineries

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Received June 6, 2017; revised and accepted August 24, 2017

View online at October 11, 2017 Wiley Online Library (wileyonlinelibrary.com);

DOI: 10.1002/bbb.1825; *Biofuels, Bioprod. Bioref.* 12:125–138 (2018)

Abstract: Pretreatment prior to or during biological conversion is required to achieve high sugar yields essential to economic production of fuels and chemicals from low cost, abundant lignocellulosic biomass. Aqueous thermochemical pretreatments achieve this performance objective from pretreatment coupled with subsequent enzymatic hydrolysis, but chemical pretreatment can also suffer from additional costs for exotic materials of construction, the need to recover or neutralize the chemicals, introduction of compounds that inhibit downstream operations, and waste disposal, as well as for the chemicals themselves. The simplicity of hydrothermal pretreatment with just hot water offers the potential to greatly improve the cost of the entire conversion process if sugar degradation during pretreatment, production of un-fermentable oligomers, and the amount of expensive enzymes needed to obtain satisfactory yields from hydrothermally pretreated solids can be reduced. Biorefinery economics would also benefit if value could be generated from lignin and other components that are currently fated to be burned for power. However, achieving these goals will no doubt require development of advanced hydrothermal pretreatment configurations. For example, passing water through a stationary bed of lignocellulosic biomass in a flowthrough configuration achieves very high yields of hemicellulose sugars, removes more than 75% of the lignin for potential valorization, and improves sugar release from the pretreated solids with lower enzyme loadings. Unfortunately, the large quantities of water needed to achieve this performance result in very dilute sugars, high energy costs for pretreatment and product recover, and large amounts of oligomers. Thus, improving our understanding of

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hydrothermal pretreatment fundamentals is needed to gain insights into R&D opportunities to improve performance, and help identify novel configurations that lower capital and operating costs and achieve higher yields. © 2017 Society of Chemical Industry and John Wiley & Sons, Ltd

Keywords: biomass; water; hydrothermal pretreatment; sugars; lignin

Introduction

With up to about 1.5 billion or more dry tons projected to be available annually at a cost of \$60/dry ton or less, lignocellulosic biomass in such forms as agricultural (e.g. corn stover) and forestry (e.g. waste wood) residues and fast growing herbaceous (e.g. switchgrass) and woody (e.g. poplar wood) energy crops represents a substantial resource.¹ In addition, lignocellulosic biomass costing ~ \$60/dry ton is competitive in price with petroleum at \$20/barrel on an equivalent energy content basis.² And these large domestic sources of inexpensive lignocellulosic biomass can be converted into liquid fuels with very low net greenhouse gas (GHG) emissions, thereby offering the potential to move the transportation sector from being the leading emitter of greenhouse gases due to its virtually complete reliance on petroleum. These unique attributes position lignocellulosic biomass as the only sustainable resource that can support large-scale, low cost production of organic fuels and chemicals.³⁻⁵ However, despite this impressive potential, deconstruction of the naturally recalcitrant complex polymers comprising lignocellulosic biomass into intermediates that lend themselves to conversion into valuable fuels and chemicals presents the major hurdle to achieving the economic viability essential to major commercial use.⁶ In the case of biological processing, the desired intermediates are the sugars that make up cellulose and hemicellulose, with enzymes generally recognized as the best option to achieve high yields of glucose from cellulose. Important factors that are credited with contributing to the recalcitrance of lignocellulosic feedstocks to biologically releasing glucose with high yields include (i) hemicellulose, lignin, and other compounds coating the surface of cellulose microfibrils; and (ii) the highly structured crystalline structure of cellulose limiting enzyme action. Although biotechnology promises to substantially reduce costs of biomass processing, pretreatment prior to or during biological deconstruction has proven vital to achieving released most of the sugars from hemicellulose and cellulose in naturally resistant lignocellulosic biomass.⁷

In this paper, pretreatment refers to disruption of lignocellulosic biomass to make cellulose and hemicellulose remaining in the pretreated solids accessible to enzymatic

hydrolysis to fermentable sugars. Over the years, many biological, chemical, and physical pretreatments have been applied to biomass in an attempt to increase cellulose susceptibility to enzymes, and several thermochemical approaches appear promising.⁸ However, although pretreatment is typically a necessary condition, it will likely not be sufficient to support economic biofuels production unless many, if not all, of the following considerations are met: low-cost or easily recovered and recycled chemicals for pretreatment and post-treatment, low-cost materials of construction through avoiding use of corrosive chemicals and high pressures, limited need for hydrolyzate conditioning with associated sugar losses prior to fermentation, minimal formation of inhibitors to the subsequent bioprocesses, limited degradation of sugars during pretreatment, low heat and power requirements, high solids loading and resulting sugar concentrations, and low waste-treatment costs. To enhance biorefinery revenues, innovative pretreatments could also recover lignin, protein, minerals, oils, and other materials contained in biomass for use as fuel, food, feed, fertilizers, chemicals, and other products. Efficient utilization of all components of biomass would also leverage the biomass resource, thereby reducing land requirements to meet product markets, a valuable outcome for a sustainable society. Regardless of the pretreatment, many forms of lignocellulosic biomass, such as agricultural residues, have high mineral salts contents (~15%) with major cations, including calcium, potassium, magnesium, sodium, alumina, and iron, many of which are considered to be bound to inorganic anions. In light of the impact on the effectiveness of chemicals employed in pretreatment,⁹⁻¹¹ mineral salts can present an important challenge to cost effectiveness. However, even if a pretreatment can meet these challenging demands and take advantage of opportunities to enhance resource utilization, a particularly critical requirement is that high sugar yields can be realized in subsequent biological operations at affordable enzyme loadings of about 2 mg enzyme protein/g biomass solids or less, as even this loading is projected to cost on the order of \$0.25/gal of ethanol made.¹²

Unfortunately, pretreatments have yet to be developed that can meet such conditions for economic viability. Thus, in light of the pivotal role of pretreatment, advanced

pretreatments that substantially reduce costs are still needed if biological processing of biomass is to become competitive for large-scale applications in an open market and thereby realize the environmental, sustainability, economic, and strategic benefits of biofuels. With numerous past trial-and-error attempts not being completely successful in this quest, understanding pretreatment mechanisms and applying that knowledge to advance pretreatment via a Pasteur's Quadrant paradigm deserves greater emphasis.¹³⁻¹⁶

Hydrothermal pretreatment attributes

A simple pretreatment that conceptually offers many attributes consistent with the goals outlined above is to heat moist plant biomass to 180–270°C with hot water or steam but without adding any other ingredients. For example, a technoeconomic model of an n^{th} plant biorefinery based on data developed by the Biomass Refining Consortium for Applied Fundamentals and Innovation (CAFI) team projected that liquid hot water pretreatment resulted in the lowest capital requirements among six pretreatments studied.¹⁷ Hydrothermal pretreatment offers the possibility of producing less inhibitory compounds that, along with chemicals used in other pretreatments, would require removal prior to downstream biological operations.^{18,19} Lower production and use of such inhibitors also results in fewer environmental penalties, including reduced greenhouse gas emissions, and improve water-quality effects from biorefineries. Yet, even though liquid hot water pretreatment had the lowest overall process capital costs of the CAFI options, the projected minimum ethanol selling price (MESP) was not the lowest as a result of sugar losses during pretreatment.¹² In particular, batch or co-current flow hydrothermal pretreatment suffers from two important challenges: (i) loss of hemicellulose sugars during pretreatment and (ii) the need for high enzyme loadings to achieve high yields in subsequent enzymatic hydrolysis of hydrothermally pretreated solids. Furthermore, hydrothermal operations do not readily recover valuable components from biomass that could enhance revenues.

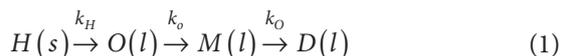
To capitalize on the key advantages of hydrothermal pretreatment, new strategies are needed to make it competitive. Macroscopic R&D can play an important role through elucidating key factors governing hydrothermal pretreatment performance, identifying mechanism responsible for key economic obstacles, and informing pathways with the potential to reduce capital and operating costs while realizing high yields from hot water pretreatment coupled with subsequent biological conversion. If suc-

cessful, innovative hydrothermal pretreatments have the potential to provide low risk and high reward platforms on which to build a major biorefining industry. In addition to being conceptually attractive due to simplicity, water only pretreatment, the focus of this paper, provides a useful baseline from which to understand and conceptualize how aqueous pretreatments that rely on chemicals alter performance and understand their advantages, pitfalls, and optimization opportunities. This paper summarizes hydrothermal pretreatment fundamentals, advantages and weaknesses of conventional batch and co-current hydrothermal pretreatment systems, and insights gained by passing hot water through lignocellulosic biomass in a flowthrough approach to illustrate some of the attributes needed for hydrothermal pretreatment to become cost competitive.

Fundamentals of hydrothermal processing effects on biomass

Hydrothermal pretreatment can be broadly classified into two technology sets based on the relationship of the reaction temperature and pressure to the critical point for water: (i) saturated steam temperatures and pressures below the critical point, i.e., subcritical water; and (ii) pressures and temperatures above the critical point, i.e., supercritical water. Subcritical water pretreatment is at temperatures ranging from 100°C to 374°C under sufficient pressure to maintain water in the liquid phase,²⁰ while supercritical pretreatments apply temperatures above 374°C with corresponding pressures of over 22.1Mpa.^{21,22} Historically, hot water pretreatment has been given a variety of labels, including liquid hot water,²³ hydrothermal,²⁴ autohydrolysis,²⁵ pH-controlled,²⁶ hydrothermolysis,²⁷ aqueous or steam/aqueous fractionation,²⁸ uncatalyzed solvolysis,²⁹ aquasolv,³⁰ subcritical water,^{31,32} and supercritical water,³¹ as well as combinations and variations of these terms. Regardless of the name, a goal of hot water pretreatment should be to increase yields to those from low pH dilute acid pretreatments while realizing lower operating and capital costs. One of the advantages hot water pretreatment offers is that hydronium ions formed by water dissociation at higher temperatures catalyzes hydrolysis of the polysaccharides in lignocellulosic biomass.³³⁻³⁵ Although water at ambient conditions has a polar nature that results in an extensive hydrogen bonding network among water molecules, heating water weakens H-bonding, and water dissociates into acidic hydronium ions (H_3O^+) and basic hydroxide ions (OH^-). These ions can then hydrolyze the polysaccharides in biomass to form sugars. This dissociation can be tuned by changing the temperature and pressure.³⁶

Hemicellulose deconstruction during hot water pretreatment can be modeled as the following series of first order reactions that are catalyzed by hydronium ions:



in which H represents hemicellulose, O refers to hemicellulose oligomers, M is the monomeric sugars released by oligomer breakdown, D refers to degradation products, (s) indicates solids, (l) refers to components in solution, and the k_i 's are rate constants for deconstruction of species i . The fermentation organisms used to biologically produce ethanol or other products from the polysaccharides in lignocellulosic biomass can usually metabolize monomeric sugars represented by M in Eqn (1) effectively but often cannot directly ferment the oligomers O. On the other hand, the series reaction depicted by Eqn (1) results in a balancing act between formation of oligomers from hemicellulose, breakdown of oligomers to monomeric sugars, and loss of sugars by degradation. Because the key to economic viability is to maximize sugar yields, the reaction time must be long enough to breakdown as much as possible of the hemicellulose to oligomers and preferably monomers while keeping it short enough to minimize the loss of sugars to degradation products that also inhibit downstream sugar fermentation. The relative values of the rate constants in Eqn (1) shift the balance to favor oligomers more than monomers, with the result that the concentration of oligomers is about 4 times (or more) higher than that of monomers at conditions that achieve high hemicellulose conversion while avoiding excessive

degradation of sugars. This balancing act is influenced by time and temperature, such that lower temperatures require longer times or vice versa to reach the same extent of reaction. The severity parameter R_0 has been applied to describe this tradeoff in conditions as calculated by the following expression:

$$R_0 = t \exp [(T-100)/14.75] \quad (2)$$

In which t is the time in minutes and T the temperature in Celsius. As shown in Fig. 1 for a conventional batch system, the severity parameter is effective in grouping combined sugar (oligomer+monomer) and oligomer yield data gathered over a range of times and temperatures. However, it is important to note that the differences in activation energies of the rate constants in Eqn (1) result in temperature influencing the reaction balance more strongly than reaction time.

The solubility of sugar oligomers and monomers released in hot water pretreatment is an important factor determining the efficiency of sugar recovery from lignocellulosic biomass.³⁷ The solvation effects of water are related to its dielectric constant (ϵ), polarity, ion product, and viscosity, which in-turn impact pH, density, and mass transfer of biomass species into solution, respectively. It was reported that higher temperatures reduced water polarity and dielectric constant (ϵ) and lower dielectric constants (ϵ) and polarities increase the solubility of organic compounds such as saccharides released from lignocellulosic biomass.³⁸⁻⁴¹ For example, while the dielectric constant of water at 25°C is approximately 81, making it an extremely

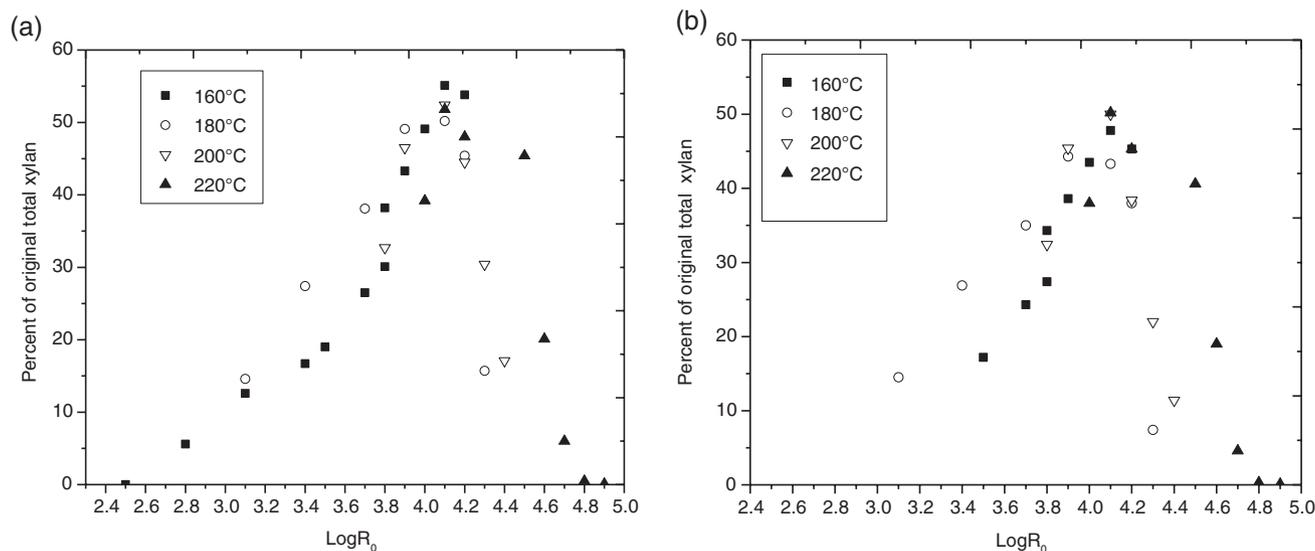


Figure 1. Combined sugar yield (a) and oligomer yield (b) in the hydrolyzate vs. $\log R_0$ for hot water batch tube pretreatment.

polar solvent, increasing the temperature to 250 °C with enough pressure applied to maintain water in the liquid state reduces the dielectric constant to approximately 27.³⁸ The latter approaches that for organic solvents such as methanol and ethanol in which organic compounds are highly soluble.³⁸ Pressure also affects the dielectric constant of water. For example, Frank (1987) showed that increasing pressure at a constant temperature enhanced the dielectric constant of water, thereby reducing its ability to dissolve organic compounds.⁴⁰ As a result, increasing pressure could reduce sugar recovery from lignocellulosic biomass during hot water pretreatment. Thus, operation at just enough pressure to maintain water in the liquid state at a given temperature, i.e., saturation conditions, should maximize sugar solubility at that temperature. Interestingly, as shown in Fig. 2, increasing the temperature and the cor-

responding pressure to the supercritical region reduces the ionization constant significantly.^{21,22} However, the high values of the water dissociation constant, K_w , at temperatures between 250 and 300°C for a pressure of 25MPa increase hydronium ion concentrations to increase catalytic efficiency potential.⁴² In addition, the viscosity and surface tension of water drop with increasing temperature while the diffusivity and self-ionization of water increase. The result is that higher temperatures increase the solubility of fragments produced by deconstruction of lignocellulosic materials by several orders of magnitude.

Water can realize several key attributes of low acid and no acid technologies.⁴⁴⁻⁴⁷ Hemicellulose, the most readily hydrolyzed constituent, is partially deacetylated, as well as depolymerized in the presence of water.²⁸ As a result, hot water pretreatment can be considered as a low pH

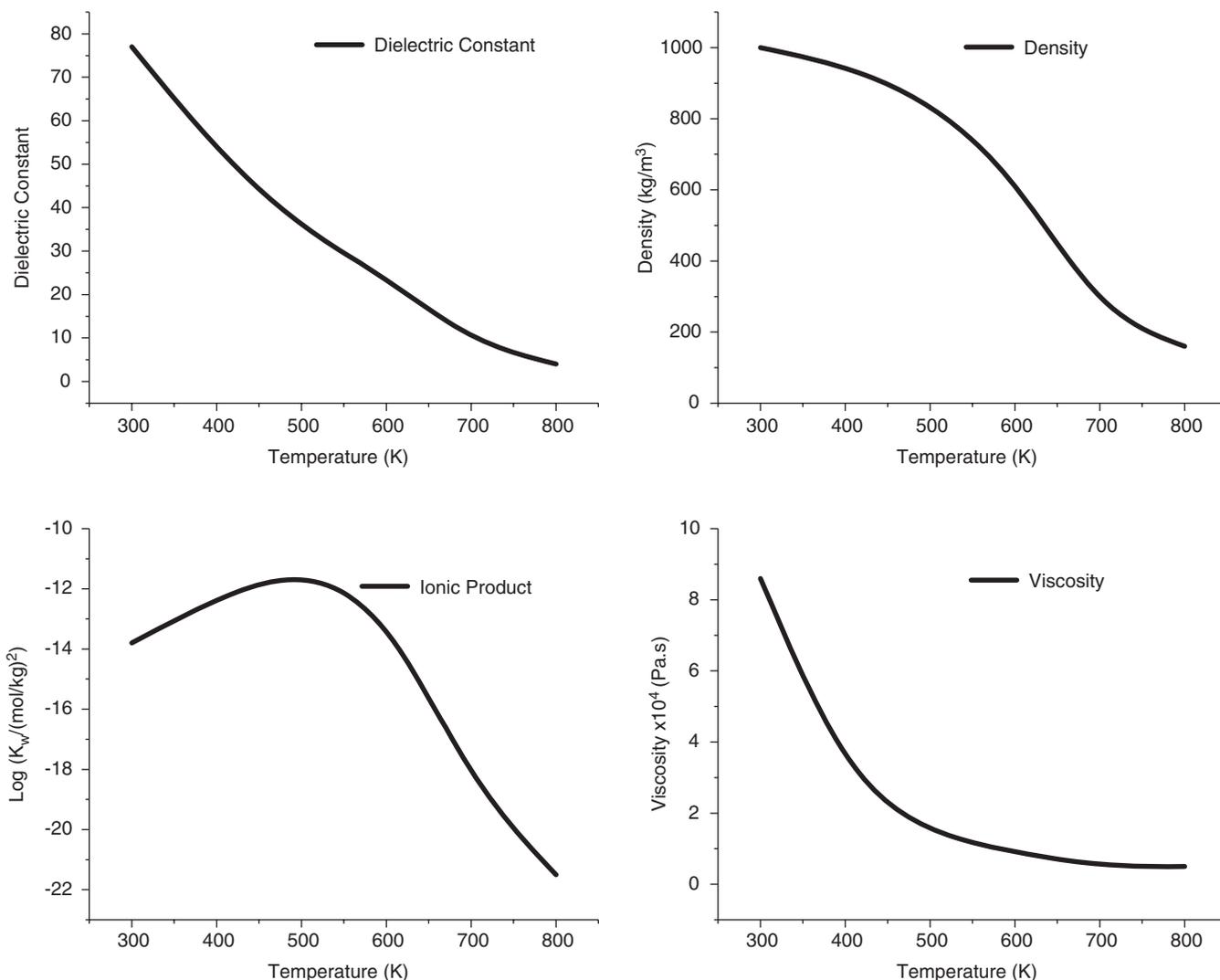


Figure 2. Changes in dielectric constant, density, ionic product, and viscosity of water with high temperatures under saturated steam pressure.⁴³

thermochemical pretreatment due to contributions of the resulting acetic and other acids released during hemicellulose hydrolysis coupled with the drop in pH of water with increasing temperature. These hydrolytic reactions also cleave some of the cellulose. Furthermore, hot water lowers the lignin softening point,⁴⁸ allowing separation of fibers⁴⁹ from internal lignin bonds and glycosidic linkages, and catalysis of hydrolytic reactions by the resulting hydrogen ions cleaves internal lignin bonds along with glycosidic linkages in both hemicellulose and cellulose to depolymerize biomass. Similarly, water can depolymerize lignin even in the absence of acetic acid or other biomass constituents.^{50,51} The increased acidity of water at elevated temperatures may enhance cleavage of chemical bonds. In addition to catalytic contributions resulting from water ionization at high temperatures, the amount of water present in pretreatment could be important in light of the low solubility of higher molecular weight oligomers even though their solubility increases with temperature.^{52,53}

As illustrated in Fig. 3, hot water pretreatment has highly desirable attributes for overcoming key biomass recalcitrance features, including (i) disrupting the cellulose/hemicellulose/lignin complex by increased removal of hemicellulose and lignin,¹⁹ (ii) lowering cellulose crystallinity to enhance the accessibility of cellulose to enzymes and increase total sugar yields,^{54–56} (iii) depolymerizing lignin to increase yields of lower molecular weight lignin fragments, and (iv) removing a large portion of lignin that otherwise hinders cellulose accessibility to enzymes and non-productively adsorbs enzymes.⁵⁷ Process conditions such as temperature and pressure, residence time, pH, biomass concentration, and water concentration likely affect the extent to which these factors are altered. In many cases, these variables must be traded-off against one another to a significant extent. For example, hot water pretreatment converts the structural components of biomass into water-soluble compounds, including

sugar oligomers and some monomeric sugars. However, monomeric sugars can degrade into furans (furfural and 5-HMF), which in turn can degrade into organic acids such as levulinic acid and further breakdown to what some call ‘humins.’ Formic acid and acetic acid are released from dissociation of hemicellulose/cellulose/lignin inter-polymeric linkages.^{58,59} And oligomers can directly degrade into various as yet not fully characterized compounds.^{18,60} Lignin is believed to depolymerize and form micelles via both homolytic and acidolytic cleavage into low molecular weight lignin globules.^{61–63}

Batch/co-current hydrothermal pretreatment

As already mentioned, the CAFI project compared the costs for batch or co-current hydrothermal pretreatments to those for dilute acid in an integrated biorefinery based on CAFI pretreatment data.^{12,65} Assuming similar pressure ratings and identical reactor configurations, the pretreatment reactor capital cost would be about 10–15% lower if stainless steel is employed for liquid hot water pretreatment instead of corrosion resistant Incoloy cladding needed for dilute acid pretreatment. In addition, eliminating the need to purchase sulfuric acid for pretreatment and ammonia for subsequent neutralization lowered the operating costs from those for the dilute acid pretreatment. In fact, hydrothermal pretreatment had the lowest projected operating and capital costs among all six pretreatments considered in the CAFI study.⁶⁶ However, although liquid hot water pretreatment had reasonably high total sugar and oligomer yields and the lowest pretreatment capital cost, its projected minimum ethanol selling price (MESP) was not the lowest because the significant fraction of oligomeric sugars released from the pretreatment were assumed to not be fermentable in the study.^{65,67,68} Although they can be hydrolyzed to monomers for fermentation,⁶⁹ the

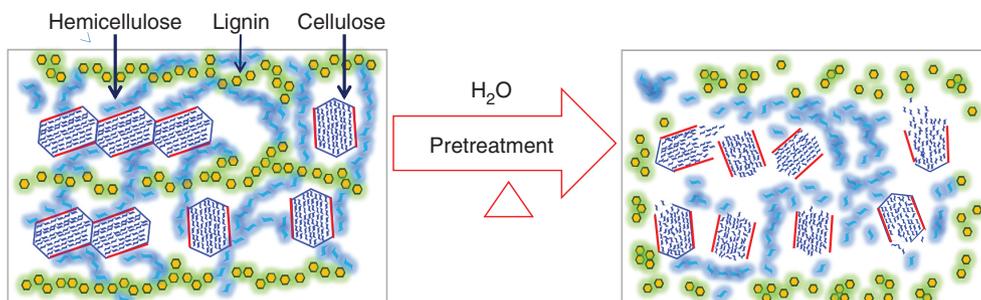


Figure 3. Conceptual illustration of the disruption of lignocellulosic biomass by hydrothermal pretreatment (modified from Zeng *et al.*⁶⁴).

extra costs would hurt the overall process economics. Nevertheless, since projected operating and capital costs of hydrothermal pretreatment were the lowest among all six pretreatments studied in the CAFI project, further research is warranted to evaluate how such extra costs affect the economics of hydrothermal pretreatment compared to other leading pretreatments.

Overall, high yields are essential to realizing low costs by spreading operating and capital costs over more products. Figure 4 presents a base case design⁷⁰ with a pretreatment yield of xylan-to-xylose of 78%, an enzymatic hydrolysis glucan-to-glucose of 81%, a fermentation yield of glucose-to-ethanol of 95.0%, xylose-to-ethanol yield of 92.6%, and arabinose-to-ethanol yield of 54.1%. If the yields can be improved, then costs can be reduced, as illustrated in Fig. 4. For instance, if pretreatment xylan-to-xylose yields were improved from the base case, i.e., 78% to 100%, then the resulting cost reduction would be 6.6% compared with the base case of Tao *et al.*⁷⁰ Figure 4 also points out that sugar yields have a stronger influence on overall costs than other parameters such as reductions in enzyme loadings. Nonetheless, process improvements can still further reduce sugar costs, particularly for pretreatment and enzymatic hydrolysis. Similarly, improved pretreatment and enzymatic hydrolysis technologies could improve the digestibility of pretreated

solids, so that yields in the biological steps are further improved. The 100% arabinan-to-arabinose yield case only improves the cost by 0.2%, mainly due to the low arabinan content in the biomass feedstock. Among the three sugars (i.e., glucose, xylose, and arabinose) considered in the 2011 NREL report, enhancing glucose yields had the greatest impact. If 95% of all three sugars were fermented to ethanol, the cost would drop by 18.8%. However, it is important to note that the values of these yield or process improvements are hypothetical and may or may not be achievable.

Comparing with acid and alkali pretreatment, elimination of the added chemicals (e.g. acids and alkalis) while achieving virtually theoretical hemicellulose sugar yields in hot water pretreatment lowered costs. Also, much less ammonia will be needed to neutralize organic acids liberated from the biomass feedstocks compared to the use of dilute acid pretreatment. Because only a small amount of ammonia is needed, neutralization with ammonia instead of lime can be more economical and environmentally friendly through eliminating generation of gypsum or other problematic residues that create operational and disposal problems.⁷¹ In light of waste-water treatment (WWT) contributing ~20% of the cost of the dilute acid process in the 2011 NREL design report, hot water pretreatment can potentially reduce WWT costs because of

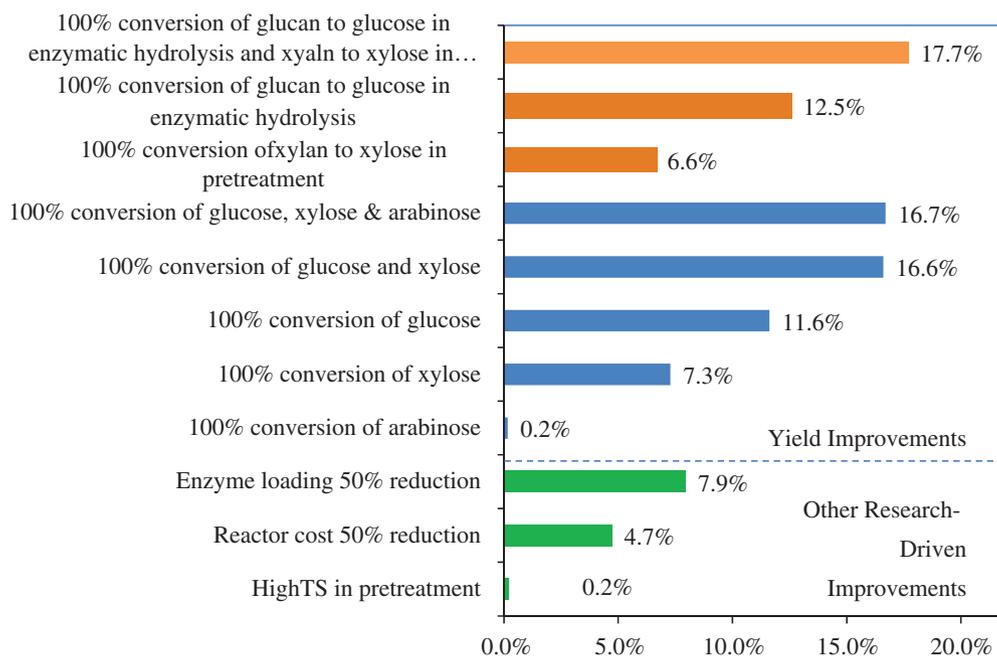


Figure 4. Economic impacts of yield improvements (orange bars for yield improvements from pretreatment and enzymatic hydrolysis and blue bars for yield improvement from biological conversion) and other research-driven improvements (e.g. hot water pretreatment with high solids loadings, lower reactor cost, or less enzyme loadings, shown as green bars).

the minimal chemicals needed upstream for hot water pretreatments. Although the extent of reduction in GHG emissions requires detailed life cycle analysis, hot water pretreatment has a potentially lower GHG footprint because energy intensive production of sulfuric acid is eliminated and less ammonia is needed.⁶⁵

Flowthrough improves hydrothermal pretreatment

Process configurations can significantly impact the performance of hot water pretreatments. Various studies have shown that countercurrent flow of liquid and solids or flow of water through a bed of solid biomass at up to about 220°C can achieve nearly theoretical hemicellulose yields,^{29,72–75} remove up to about 70% of the lignin and 20% of cellulose, produce highly digestible cellulose in the remaining solids^{8,76} (Fig. 5), and produce liquid hydrolyzate that is more compatible with fermentative organisms.⁷⁷ Furthermore, application of much higher temperatures of about 225–270°C can increase lignin removal to about 95% and completely solubilize crystalline cellulose, as shown in Fig. 5.^{32,78,79} Such hot water flowthrough systems quickly carry solubilized materials from the reaction zone, thereby limiting the opportunity for degradation. However, removal of components soon after they are dissolved results in a very high fraction of oligomers, thereby requiring introduction of a downstream oligomer hydrolysis operation if fermentative organisms cannot metabolize

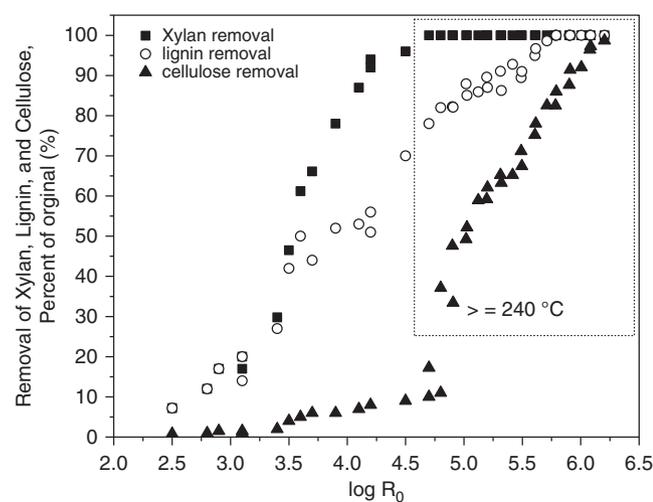


Figure 5. Effect of pretreatment severity on xylan, lignin, and cellulose removal from poplar wood by hydrothermal flowthrough pretreatment over the temperature range of 160–280°C and reaction times of 0–60 min for a water flow rate of 25 mL/min.³²

such long chained polysaccharides.^{32,63,78,81} Flowthrough pretreatments at about 200°C with very dilute sulfuric acid (e.g. 0.05% H₂SO₄) show similar results to those for flowthrough with just hot water, with the exception that more cellulose is solubilized and monomeric sugar yields are higher.²⁵

As early as the 1970s and 1980s, Bobleter *et al.*⁸² applied flowthrough hot water to hydrolyze air-dried pure cellulose at 260–270°C and achieve up to 52% glucose and 10% 5-hydroxymethylfurfural (5-HMF) yields at 265°C with a flow rate of 12 mL/min. Furthermore, a two-stage semi-flow hot water pretreatment (230°C for 15 min and 270°C for 15 min) at a flow rate of 10 mL/min at a pressure of 10 Mpa removed 100% of xylan, 89.4% of Klason lignin, and 79.5% of cellulose. However, substantial sugar degradation to furfural, 5-HMF, glycolaldehyde, and other products resulted,⁸³ along with high distributions of oligomers and low concentrations of degradation products.^{18,19} Furthermore, a sizable portion of hemicellulosic and lignin sheath surrounding cellulose microfibrils was rapidly solubilized and removed from the reaction zone, and the resulting increased biomass porosity and enzyme accessibility to cellulose microfibers increased sugar yields at lower enzyme loadings in subsequent biological hydrolysis. If the hot stream was flash cooled, lignin deposited in the flash vessel while solubilized hemicellulosic sugars remained in the flash cooled liquor.⁸⁴ These results contrast with those from batch systems in which solubilized lignin deposits on pretreated lignocellulosic biomass solids to hinder enzyme access and reduce enzymatic saccharification yields.

Hydrolyzates from most pretreatments require conditioning prior to subsequent fermentation to remove inhibitors released from natural biomass ingredients (e.g. acetic acid) or formed through biomass degradation (e.g. furfural and lignin degradation products).^{85,86} Pretreatment with just water minimizes oligosaccharide hydrolysis, while hydrating their structure by liquid water above the saturation vapor pressure of water at the operational temperature.⁸ However, the high fraction of oligomers reduces the opportunity for monosaccharides to rapidly degrade to aldehydes during high temperature pretreatment, thereby increasing soluble saccharide yields. Even if some of the hemicellulose sugars are left in the solids for conditions that keep sugar degradation low, the enzymes used to release glucose from the cellulose-enriched solids produced by hydrothermal pretreatment also solubilize a high portion of the hemicellulose. As an example, hot water pretreatment of corn fiber from corn wet milling combined with subsequent enzymatic hydrolysis led to nearly

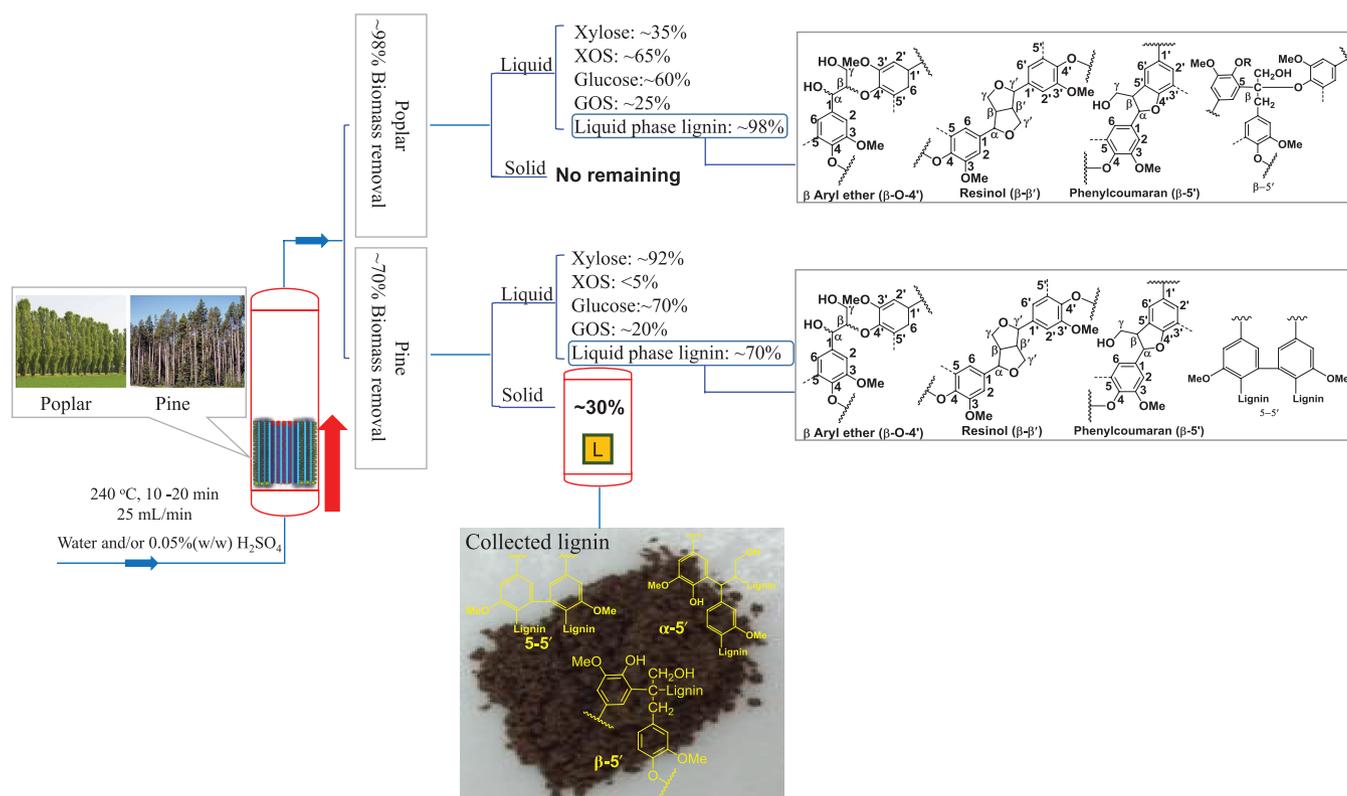


Figure 6. Schematic of water-only and dilute acid flowthrough pretreatment of biomass along with picture of lignin solids left after hydrothermal pretreatment.^{32,62,63,78}

complete recovery of cellulose and hemicellulose sugars as well as starch in pretreated residues.⁸⁷

Increasing hot water flowthrough pretreatment temperatures to 225–270 °C while maintaining pressure at or above the saturated steam pressure solubilizes more biomass^{54–56} but also significantly increases biomass degradation. The hemicellulose in lignocellulosic biomass is particularly prone to degrade at high temperatures; for example, hydrothermal operation at temperatures of 180 to 240 °C can easily decompose most of the hemicellulose. Nevertheless, as shown in Fig. 5, the severity parameter R_0 can be carefully optimized to minimize formation of furfural and other degradation products that hurt yields and inhibit fermentations.⁸⁸ For this reason, the residence time is usually varied over a range of reaction temperatures (typically 180 to 240 °C) and times to determine the combination that maximizes sugar plus oligomer yields from pretreatment coupled with subsequent enzymatic hydrolysis.⁸⁹

Our laboratory research with flowthrough pretreatment of poplar wood pictured in Fig. 6 at elevated temperatures (200–280 °C) revealed that hydrothermal pretreatment above 240 °C or with 0.05% (w/w) H_2SO_4 at 220 °C signifi-

cantly disrupted and removed cellulose and resulted in more than 98% total biomass removal.³² The result was up to 100% of the xylan and 90% of cellulose were solubilized with negligible furfural and 5-HMF formation during pretreatment. It was found that xylan was predominately removed as soluble xylooligomers with some xylose, about 86% of the glucan removed was as soluble glucose oligomers and glucose, and cellulases readily digested the remaining glucan/cellulose to obtain a nearly 100% yield.^{32,90} Most of the lignin removed precipitated when the liquid hydrolyzate from pretreatment was cooled.³² Modest structural changes of lignin side chains were observed, with formation of benzylic carbonyl groups at the α -position, α - β unsaturation, and oxidation of the γ -OH to aldehydes. Formation of these propenyl end groups during pretreatment was plausibly due to cleavage of β -O-4' aryl ether by dehydration at the α -position and oxidation at the γ -OH position. In addition, linkages in insoluble lignin, such as resinol, β -O-4' and the phenylcoumaran structures, were observed, and slight repolymerization formed new C β -C5' linkages in the insoluble lignin.⁶³ For hardwood lignin, slight condensation of C β -C5 and loss of γ -methyl groups occurred in the lignin

fraction removed.⁸⁰ While hardwood lignin was completely removed into the hydrolyzate, ~30 % of softwood lignin remained as solid residues under identical pretreatment conditions, plausibly due to vigorous C5-active recondensation reactions (C-C5). The low molecular weight lignin that precipitated from the hydrolyzate could be catalytic upgraded to C7 to C18 range hydrocarbons through recent advances in hydrodeoxygenation.⁹¹⁻⁹³

Advancing hydrothermal pretreatment to reduce costs

Flowthrough hot water pretreatments are distinctive from seemingly similar batch steam or hot water systems with or without acid addition in that they achieve higher sugar and lignin yields. Unfortunately, the large amount of water needed for flowthrough to realize such results significantly dilutes sugar streams, resulting in high-energy requirements for both pretreatment and product recovery as well as larger fermentation vessels. In addition, the high pressures required for pushing water through biomass increase containment costs and challenges. Thus, laboratory configurations would not appear to be commercially viable. Nonetheless, due to their unique and powerful attributes, successful development of new hot water flowthrough pretreatments could prove vital to low cost biological production of ethanol from biomass.⁹⁴ To realize this potential, flowthrough processes must reduce energy demands and costs for fermentation and product recovery. Large-scale commercial devices that pass hot water through wood chips have achieved success but appear to be less plausible for application to lignocellulosic biomass materials such as switchgrass or corn stover that pack together too tightly to allow water to easily move through them (<https://www.andritz.com>).

Continuous flowthrough hot water pretreatment faces several technical and economic challenges. First, hot water needs to flow through or counter-current to the biomass. Second, the total solids loading needs to be high to produce high sugar concentrations. A single stage counter current reactor is desired in that two-stage counter-current reactors suffer from loss of biomass structure in the transfer between stages while operating counter current pretreatment and extraction on mud like particles would be difficult.⁶⁸ Inclined screw extractors may be modified to allow diffusion of dissolved biomass components into the bulk flowing stream for rapid removal from the reaction zone if they can be made to operate at the elevated temperatures and pressures for hot water pretreatments. On the other hand, low total solids loading in pretreatment not only adds

more water, thus increasing the size of the pretreatment reactor and downstream equipment, but also increases the amount of energy required to remove the product from the water. When the total solids loading is increased from 30% to 35%, the minimum ethanol selling price of \$2.15/gallon (2007\$) predicted by the NREL 2012 lignocellulosic design to be cost-competitive with other transportation fuels⁷⁰ dropped by less than half of a percent. However, when total solids loading is reduced below 15%, imported energy (natural gas or electricity) would be needed because the energy generated from burning lignin and residual sugar is insufficient to remove ethanol product from the water as well as meet all other biorefinery energy demands.

Although flowthrough technology may not be commercially viable on all feedstocks without significant equipment breakthroughs, its operation can provide valuable insights into biomass deconstruction. Hot water flowthrough pretreatment can achieve similarly high sugar yields to dilute acid pretreatment and is very effective at high lignin solubilization.^{32,92} In addition, hot water also extracts soluble metal ions Na, K, P, and nitrogenous compounds (e.g. proteins) that could have value as potassium and phosphorous rich fertilizer. Removal of these soluble components from the residual solids during pretreatment will lower boiler operating costs by reducing slagging, attack of the refractory material by these compounds, and NOx emissions.

Conversion of all major biopolymers within biomass, including lignin, provides a major opportunity for realizing low costs, thereby enhancing the overall operational efficiency, economic viability, and sustainability of biofuels production. However, despite its potential importance, lignin conversion to biofuels or other valuable commodity products has proven to be elusive.^{91,95-100} Thus, it is important to reinvestigate development of innovative pretreatments that support lignin valorization while still realizing high product yields from structural carbohydrates at low enzyme loadings are needed to enhance the economic viability of lignocellulosic biomass refineries.⁷

Closing thoughts

Conventional batch or co-current hydrothermal pretreatment offers six important cost advantages: (i) minimal chemical costs, (ii) ability to handle large biomass particles, (iii) reduced production of biological inhibitors, (iv) relatively low-cost containment, (v) the ability to process high solids concentrations, and (vi) short residence times. However, sugar yields from hemicellulose are low, and high loadings of expensive enzyme must be applied to

achieve high glucose yields from hydrothermally pretreated solids. In addition, the lignin produced does not lend itself to transformation into valuable products. Furthermore, although hot water pretreatments currently require high temperatures and attendant high pressures to be most effective, operation at lower operating pressures would reduce capital and energy costs for feeding and discharging biomass as well as maintenance. Thus, advances in hydrothermal pretreatment technology are needed to maintain the advantages of current batch/co-current systems while enhancing sugar yields during pretreatment and supporting hydrolysis of polysaccharides left in pretreated solids with low enzyme loadings.

Continual flow of hot water through lignocellulosic biomass that is either stationary or moving countercurrent to the flow of water shows that it is possible to enhance the performance of hot water pretreatment. In particular, hemicellulose sugar yields can be substantially increased, albeit mostly as oligomers that likely require further processing to be fermentable. Such flowthrough operations also remove a large portion of lignin, thereby potentially enhancing overall biorefinery revenues. Although commercial flowthrough systems have been applied to hydrothermally pretreat wood chips, high loadings of expensive enzymes are still required to realize commercially viable sugar yields from the solids produced by laboratory and large-scale systems. Furthermore, the large amounts of water applied in laboratory flowthrough pretreatments to achieve these outcomes results in excessive energy demands for pretreatment and product recovery. Thus, novel process configurations are needed to meet the full spectrum of key performance metrics from pretreatment with just hot water.

Nonetheless, flowthrough operations provide valuable insights into the fundamentals of hydrothermal pretreatment and suggests routes achieve some important advantages. Improved knowledge of the relationship between pretreatment conditions, process configurations, and controlling mechanisms is vital to selecting appropriate commercial equipment and realizing targeted total sugar and lignin solubilization in commercial operations. In this context, a better understanding of aqueous biomass interactions at operational temperatures and pressures, especially in terms of the catalytic and solvation effects of water and biomass reaction kinetics, mass transfer, and solubility would be of great value. Development of such an understanding based on fundamental principles would also greatly aid in designing new pretreatment configurations and reducing the risk of their scale up from pilot to commercial operations. In the absence of such knowledge,

pretreatment coupled with enzymatic hydrolysis must be successively demonstrated over a wide range of scales to establish yields of hemicellulose and cellulose sugars from these operations combined before commercialization is possible, a timely and costly undertaking. In addition, limitations in our current understanding slow rational advancement of pretreatments to realize lower costs and better performance.

Acknowledgements

Dr Yang is grateful to the US Department of Energy (DOE) Award # DE-EE0006112 with the Bioproducts, Science & Engineering Laboratory and Department of Biological Systems Engineering at Washington State University for funding this research. Dr Wyman acknowledges valuable support by the Office of Biological and Environmental Research in the Department of Energy (DOE) Office of Science through the BioEnergy Science Center (BESC) at Oak Ridge National Laboratory (Contract DE-PS02-06ER64304) and the DOE BioEnergy Technology Office (BETO). He also recognizes important support by the US Department of Agriculture National Institute for Food and Agriculture (USDA NIFA). Dr Wyman also acknowledges the Ford Motor Company for funding the Chair in Environmental Engineering that facilitates projects such as this one and the Center for Environmental Research and Technology (CE-CERT) of the Bourns College of Engineering for providing facilities. In addition, the authors thank Dr Melvin Tucker from the National Renewable Energy Laboratory for insightful discussions and Dr Lishi Yan from the Suzhou University of Science and Technology who provided several figures and data.

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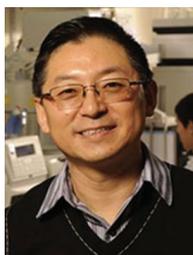
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