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Microwave pretreatment for enzymatic saccharification of sweet sorghum bagasse

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ABSTRACT

Pretreatment of sweet sorghum bagasse (SSB) through microwave radiation was evaluated at four lime doses (0, 0.1, 0.15, and 0.2 g/g SSB), two water content of 10 or 20 ml/g SSB, and three exposure times as 2, 4, and 6 min. Optimal pretreatment condition was identified as 0.1 g lime and 10 ml water per g SSB in 4 min. Under this condition, sugar yield of 32.2 g/100 g SSB (equivalent to 52.6% of maximal potential sugars) was achieved. With the same water content and exposure time, but without lime, sugar yield of 39.8 g/100 g SSB (equivalent to 65.1% of maximal total sugars) was observed. The higher sugar recovery without lime was mainly due to high sugar release during pretreatment. But with lime, sugar degradation took place, which resulted in less sugar yield though lime did make cellulose more accessible to enzymes as evidenced by higher percentage of increase of total reducing sugars during enzymatic hydrolysis. Results from this study were strongly supported by FTIR and SEM images. Overall, in a very short time and simple setup, microwave radiation shows great promise to be a leading pretreatment technique for SSB.

1. Introduction

Lignocellulosic materials such as agricultural waste and energy crops are one of the major renewable resources for fuels and chemicals. Biochemical conversion of lignocellulosic biomass to fuel ethanol requires four major unit operations, namely, pretreatment, hydrolysis, fermentation, and product separation/purification [1]. Among these, the first two are major bottlenecks in efficient conversion to ethanol. Therefore, major breakthroughs in these two operations are required to make the biochemical conversion process economical at commercial scales. Lignocellulosic biomass is composed of cellulose fibrils embedded in a less well-organized hemicellulose matrix which, in turn, is surrounded by an outer layer of lignin. The proportions of these structural carbohydrates vary by plant species, growth conditions and plant parts.

Sweet sorghum is a drought tolerant crop that requires lower inputs costs and grows in marginal lands. It is more efficient than competing lignocellulosic biomass feedstocks for bio-ethanol production [2]. It requires only 25% of the water needed by sugarcane and its cost of production is also

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one fourth of sugarcane [3]. Sorghum is grown on more than 42 million hectares in 99 countries, among which the US is the topmost producer followed by Nigeria, India and China [4]. One acre of sweet sorghum yields 30–40 tons of crops that can be processed into about 8000 gallons of sugar juice yielding about 400 gallons of ethanol. It can also produce about 10–12 tons of bagasse (crushed stalk) that can be converted into cellulosic ethanol generating about another 400 gallons of ethanol. Furthermore, as a lignocellulosic feedstock, the cell walls of sweet sorghum stems are already broken during the juice extraction step. The resulting bagasse will thus probably require milder pretreatment techniques than those for other dedicated energy crops and stover.

Several research groups have already reported their work on sweet sorghum bagasse (SSB) pretreatment, such as sulfuric acid [5,6], hydrochloric acid [7], phosphoric acid [8], steam [9], dilute ammonia hydroxide [10], ammonia fiber explosion [11], and hot water [12]. However, little comparative data are available on these promising pretreatment techniques due to different sources of sorghum, different supplies of cellulase enzymes, and dissimilar analytical methods. Thus, there is still a need to find suitable pretreatment strategies for improving enzymatic saccharification of SSB from local sweet sorghum farms.

Microwave irradiation is a promising technology that offers faster processing due to rapid and efficient heating and increased reaction rates. It has been widely used in several areas such as chemical synthesis [13], solvent extraction [14], and solid state reactions [15]. Commercially, microwave processing is successfully adopted by several industries such as food processing, composite manufacturing, and solid waste incineration, as indicated by several manufacturers of commercial scale microwave processing systems such as Ferrite Microwave Technologies, Nashua, NH (www.ferriteinc.com); and SAIREM, France (www.sairem.com). Microwave radiation corresponds to a frequency range of 300 MHz–30 GHz of electromagnetic radiation. To prevent any interference with RADAR telecommunication, microwave ovens operate at 915 MHz (industrial) and 2450 MHz (domestic). The interaction of microwave with lignocellulosic biomass causes mechanical and thermal changes on the structure of biomass. Microwave causes vibration of polar bonds inside biomass as they align themselves with the magnetic field of microwave. This causes disruption and shock to the polar bonds which accelerates chemical, physical and biological reactions [16]. The dielectric properties of materials dictate the behavior of the material when subjected to microwave electric fields. The absorption of microwave power, P (W/m^2) propagating through a dielectric material can be expressed as

$$P = 55.63 \cdot 10^{-12} f E^2 \varepsilon'' \quad (1)$$

where E represents the root mean square (rms) of electric field intensity in V/m; f is frequency of microwave in Hz; and ε'' is the dielectric loss factor, which includes the energy losses in the dielectric material due to all operating dielectric relaxation mechanisms and ionic conduction. From Eq. (1) we can see that the microwave power absorption by a dielectric material is directly proportional to the frequency of microwave, dielectric loss factor, and square of the microwave electric field intensity. Thus the intensity of microwave induced changes in biomass depends not only on the electric field intensity but also on the dielectric properties of biomass and surrounding medium (such as solvents), subjected to a particular frequency of the microwave [17].

Two studies in the 1980s were reported using microwave pretreatment for preparing cellulosic materials for downstream processes [18,19]. Recent studies on microwave pretreatment for lignocellulosic biomass include corn stover [20], wheat straw [21], rice straw [22], soy hull [23], rice hull [24], and switchgrass [25,26]. Though operating parameters regarding power level, sample residence time, and temperature are different among all of those studies, the general conclusion is that microwave is effective in disrupting the rigid lignocellulosic structures and assisting reducing sugar release. However, with regard to the effects from the solution that the samples are soaked in before microwave, either water, acid, or alkali, the results vary significantly for different biomass feedstocks. In addition, though sodium hydroxide is tested in microwave by several research groups, only one study has examined lime in microwave in case of pretreatment of wheat straw [21]. Compared with sodium hydroxide, lime is inexpensive, safer to use and easier for recovery. Lime itself has been successfully used for pretreatment of corn stover and sugarcane bagasse [27–29]. Therefore, in this study, we focus on using lime as a cheap source of alkali to assist microwave pretreatment of SSB.

Though microwave radiation has been used for pretreating different biomass feedstocks as listed above, no study has ever tried microwave on SSB to the best of our knowledge. As revealed by different investigations [26–29], the optimal pretreatment condition for achieving maximal sugar release is significantly different for different biomass materials. While with microwave radiation, alkali has been reported to enhance yield of reducing sugars [25] and is better than dilute sulfuric acid for switchgrass [26], it is not as good as dilute sulfuric acid for wheat straw [21]. Therefore, for any biomass that is dedicated for producing biofuels, the best condition for pretreatment needs to be elucidated specifically for that feedstock. This study serves the purpose of evaluating whether lime-assisted microwave pretreatment is appropriate for SSB from local farms.

The objectives of this study were to: (1) assess the effect of microwave pretreatment with or without lime on SSB structure and components; (2) identify the optimal parameters for pretreatment based on fermentable sugar release during enzymatic saccharification of pretreated SSB; (3) determine the optimal enzyme dose for hydrolysis; and (4) observe structural changes caused by microwave through fourier transform infrared spectroscopy (FTIR) and scanning electron microscopy (SEM).

2. Materials and methods

2.1. Biomass

Freshly expressed SSB was collected from Sorghum Ridge Farms, Cobden, IL, USA, washed with water, and dried at 50 °C in a hot air dryer. SSB was ground by a cutting mill (Thomas