Songklanakarin J. Sci. Technol.
41 (1), 237-245, Jan. - Feb. 2019

http://www.sjst.psu.ac.th

Original Article

Optimization of soluble sugar production from pineapple peel by microwave-assisted water pretreatment

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Received: 15 April 2017; Revised: 18 October 2017; Accepted: 24 October 2017

Abstract

The purpose of this study was to assess the potential of pineapple peel as a feedstock for fermentable sugar production. Soluble sugar production from pineapple peel was conducted in a process involving microwave-assisted water pretreatment without a catalyst in an apparatus set which prevented any loss of liquid or corrosion from vapor and reduced energy consumption. The pretreatment variables consisting of biomass loading (100-160 g/L), microwave power (90-900 watts) and irradiation time (5-20 minutes) were investigated by response surface methodology. The maximal total sugar yield in the liquid fraction after the pretreatment was 80.2% (80.2 g total sugars per 100 g dried peel) obtained using 100 g/L biomass loading at 900 watts for 9 minutes. Under these conditions, the glucose yield was 7.8%. This process offers an alternative approach to the cost-effective production of fermentable sugars from biomass-waste products, using less reagent and low energy in a self-reliant technology.

Keywords: pineapple peel, microwave-assisted water pretreatment, thermal treatment, fermentable sugars, soluble sugars

1. Introduction

A potential source for the production of bioethanol is lignocellulosic biomass, widely available in the form of agricultural and industrial wastes/residues (Limayem & Ricke, 2012). In tropical countries, the peels or skins of fruits produced as wastes/residues from the fruit canning industry constitute potential alternative feedstocks for the large-scale production of bioethanol (Arapoglou, Varzakas, Vlyssides, & Israilides, 2010; Oberoi, Vadlani, Saida, Bansal, & Hughes, 2011; Su, Tzeng, & Shyu, 2010). The world's largest producer of pineapples is Thailand with about a 17% share of the world market, and more than 40% of its pineapple exports are in canned form (Holt, 2010). The pineapple is a tropical plant whose fruits can be consumed fresh, or in cooked, juiced or preserved form and are a good source of both vitamin C and manganese. The peels/skins make up 35-50% of the pineapple fruit (Abdullah & Mat, 2008) by fresh weight, and contain comparatively high concentrations of sucrose, glucose, fructose and other nutrients (Hamalatha & Anbuselvi, 2013) that are useful for ethanol fermentation.

The typical conversion of lignocellulosic biomass to bioethanol involves three main steps, pretreatment, hydrolysis and fermentation (Balat, Balat, & Cahide, 2008). Pretreatment removes the unwanted components of lignocellulose, and alternative physical, chemical and biological pretreatments have been studied (Alvira, Tomas-Pejo, Ballesteros, & Negro, 2010; Mohan, Kumar, & Reddy, 2013; Ruangmee & Sangwichien, 2013). The crucial hydrolysis step produces fermentable sugars, and chemicals and enzymes are widely used in this relatively slow and expensive step (Demirbas, 2011).

However, the thermal treatment of lignocellulosic biomass by microwave heating can be fast and efficient (Chen, Tu, & Sheen, 2011), and has been employed in the pretreatment and/or the hydrolysis of rice straw (Gong, Liu, & Huang, 2010), wheat straw (Xu *et al.*, 2011), rape straw (Lu, Xi, & Zhang, 2011), sorghum (Chen, Boldor, Aita, & Walker, 2012), corn stover (Li & Xu, 2013) and sago pith waste

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(Thangavelu, Ahmed, & Ani, 2014). Microwave pretreatment at low temperature and pressure can be both energy efficient and economical (Boonmanumsin *et al.*, 2012; Kannan, Ahmed, & Ani, 2013; Tsubaki, Oono, Onda, Yanagisawa, & Azuma, 2012). In general, microwave-based technologies are easy to operate and tend to reduce processing energy requirements, thus enabling selective processing (Datta, 2001; Gabriel, Gabriel, Grant, Halstead, & Mingos, 1998).

The purpose of this study was to add value to pineapple crops by utilizing their peel and to assess the potential of pineapple peel as a feedstock for the production of sugar before ethanol fermentation. The study investigated an alternative process offering a cost-effective approach to the production of fermentable sugars. The process involved the water pretreatment of the pineapple peel without a catalyst/accelerator to produce soluble sugars using microwave heating. Response surface methodology (RSM) was employed to analyze the effects on the reducing sugar and total sugar yields of varying the biomass loading, microwave power and irradiation time.

An added benefit from the use of pineapple peel is that the solid fraction remaining after completing the process described in this work, can be used as the raw material for the production of further fermentable sugars and ethanol in subsequent processes (not described in this work). Furthermore, sugar production, the most important step in producing renewable energy utilizing agricultural waste was conducted in a process requiring less reagent, low energy and a self-reliant technology in keeping with the concept of progress with self-sufficiency advocated by His Majesty the late King Bhumibol Adulyadej.

2. Materials and Methods

2.1 Materials

Pineapple peel was collected from a fresh fruit market in Hatyai, Songkhla province, Thailand. The composition of the whole pineapple peel used in this study (Table 1) was determined by the Agro-Industry Development Center for Export (ADCET), Faculty of Agro-Industry, Prince of Songkla University.

Table 1. Composition of pineapple peel before and after microwave-assisted water pretreatment with 100 g/L biomass loading at 900 watts for 9 minutes.

Component		% dry weight	
		Raw peel	Solid fraction after treatment
Indigestible carbohydrate	Cellulose	33.9	41.8
	Hemicellulose	16.1	20.3
	Lignin	11.4	10.3
Digestible carbohydrate	Starch	22.8	13.7
	Total sugar	3.7	0.6
Others		12.1	13.3

The whole peel was dried using a hot air oven at 45 °C until it reached 10% w/w moisture content. Then it was crushed in a domestic blender to an average size of 2 mm which is the optimal biomass size for physical pretreatment (Sun & Cheng, 2002). The reduction in size of biomass increases the accessible surface area improving the efficiency of microwave treatment (Zheng, Pan, & Zhang, 2009). The dried-crushed peel was packed in sealed plastic bags and stored at room temperature until use.

2.2 Soluble sugar production by microwave pretreatment

The dried-crushed peel was pretreated using the microwave unit illustrated in Figure 1. The pretreatment was implemented in a 2-L glass three-necked round-bottom flask placed in a domestic microwave oven (Samsung MW71C, 2450 MHz). The top wall of the microwave oven was drilled to accommodate the flask-necks, with any gap between the oven wall and the flask-necks being covered with aluminum foil to prevent exposure to microwave radiation. Two necks of the flask were connected to a reflux glass-condenser set, one neck being attached to a tube allowing water vapor to enter the condenser and the other allowing the condensate to flow back into the flask. The third flask-neck was not used and was sealed with a cap.

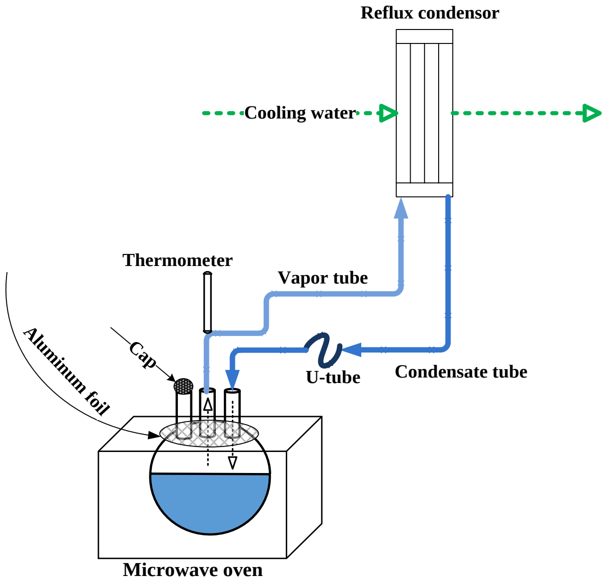

Figure 1. A schematic diagram of the microwave unit for soluble sugar production by microwave pretreatment

The experimental conditions for the microwave pretreatment were varied as presented in Table 2. The peel was mixed with 1 L deionized water in the flask, at various weights of dried peel (100-160 g), and various microwave power settings (90-900 watts) were used for 5-20 minutes irradiation times. The time durations employed were in the appropriate range of time for efficient microwave heating (Xu *et al.*, 2011).

Table 2. Codes and actual levels of the process variables for the experiment design using central composite design.

Process Variables	Symbols	Coded levels				
		-1	-0.595	0	0.595	1
Biomass loading (g/L)	B	100	112	130	148	160
Microwave power (watts)	W	90	270	450	720	900
Irradiation time (min)	I	5	8	13	17	20

As the peel and water mixture was boiled by microwave heating, water vapor rose up the condenser column in which the vapor condensed into its liquid phase releasing thermal energy which was removed with cooling water flowing through the column. The cooled liquid (condensate) then flowed back down into the mixture below via the condensate tube which was fitted with a U-shaped tube, holding liquid condensate, thus preventing any vapor reentering the flask while allowing the condensate to flow back unimpeded. The upward flow of vapor was detected by rising temperatures measured by a thermometer installed on the vapor tube.

Although the operation unit itself was under external atmospheric pressure with the end of the condenser column open to the atmosphere, the mixture in the flask was in a partial vacuum so the liquid had a lower boiling point than at atmospheric pressure. Once a steady state was reached, vaporization and condensation continued at a saturated temperature of the mixture of 97 °C under its own pressure in the flask. Variation of the microwave power influenced the time required to reach a steady state at a constant temperature of 97 °C, and the time/power relationship is as shown in Table 4. The pretreated product was filtered to obtain the liquid fraction for the analysis of its reducing sugar and total sugar contents.

2.3 Analytical methods

The solid fraction remaining after filtering out the liquid fraction produced under the near optimal conditions was analyzed according to AOAC methods (Association of Official Analytical Chemists [AOAC], 1990) and its composition is shown in Table 1. Meanwhile, the liquid fraction was analyzed for its reducing sugar content by the DNS method (Miller, 1959) and the total sugar content by the modified phenol sulfuric method (Dubois *et al*., 1956), using a UV-Vis spectrophotometer (UV, HP8453 with Chem-Station software). The reducing sugar yield and the total sugar yield were calculated by Equation (1).

Reducing sugar or total sugar yield (%)

$$= \frac{\text{sugar in liquid fraction (g/L)} \times \text{liquid fraction volume (L)}}{\text{dried peel (g)}} \times 100\% \qquad (1)$$

2.4 Response surface methodology (RSM)

The responses based on the reducing sugar and total sugar yields were studied. A central composite design (CCD) for three process variables of circumscribed type provided 14 non-repeated experiments and 3 replicates at the central level of each variable (17 total experiments). The three variables (biomass loading, microwave power and time) at different coded and actual levels are presented in Table 2, and the design conditions are shown in Table 3.

Table 3. The CCD experimental conditions and experimental results for the soluble sugar production using microwave-assisted water pretreatment.

Experiment number	CCD experimental conditions			Experimental results	
	Biomass loading (g/L)	Microwave power (watts)	Time (minutes)	Reducing sugar yield (%)	Total sugar yield (%)
1	0(130)	0(450)	0(13)	6.5	45.4
2	0(130)	0(450)	1(20)	3.5	31.4
3	0.595(148)	-0.595(270)	0.595(17)	5.3	36.5
4	1(160)	0(450)	0(13)	4.1	35.2
5	0(130)	0(450)	0(13)	6.5	45.3
6	-0.595(112)	0.595(720)	0.595(17)	3.5	49.3
7	0.595(148)	0.595(720)	-0.595(8)	6.0	27.3
8	-1(100)	0(450)	0(13)	3.8	76.0
9	0.595(148)	-0.595(270)	-0.595(8)	6.1	32.4
10	0(130)	-1(90)	0(13)	7.8	46.8
11	0(130)	1(900)	0(13)	5.2	41.1
12	-0.595(112)	-0.595(270)	0.595(17)	6.6	57.3
13	0(130)	0(450)	-1(5)	5.5	31.4
14	-0.595(112)	0.595(720)	-0.595(8)	4.7	63.3
15	0.595(148)	0.595(720)	0.595(17)	4.0	28.2
16	0(130)	0(450)	0(13)	6.4	45.3
17	-0.595(112)	-0.595(270)	-0.595(8)	6.6	53.1

Table 4. Heating time necessary to reach a steady state at a temperature of 97 °C using the microwave unit.

Microwave power (watts)	Time to reach a steady state (min)
90	50
270	18
450	11
720	8
900	6

3. Results and Discussion

3.1 Pineapple peel composition

As shown in Table 1, the major components of the solid peels on a dry basis before and after microwave pretreatment are cellulose, hemicellulose and starch (polysaccharides), which can be hydrolyzed to fermentable sugars. This indicates that the pineapple peel, which is a waste, widely produced and available from canning industries in Thailand, may be an appropriate feedstock for the commercial production of ethanol.

The decrease in the starch and lignin contents after pretreatment resulted in an increase in the cellulose and hemicellulose contents, implying that part of the starch can be degraded and the lignin structure partially disrupted using microwave-assisted water pretreatment without using any catalyst/ accelerator/ enzyme. Mainly, microwave-assisted alkali/acid pretreatment of lignocellulosic biomass is used to remove lignin, which increases the accessible surface of the cellulose/hemicellulose (Singh & Bishnoi, 2012; Suet-Pin *et al.*, 2014), and microwave hydrolysis of cellulose/starch can also be used to produce fermentable sugars (Hermiati, Mangunwidjaja, Sunarti, Suparno, & Prasatya, 2010).

This study focused on the use of microwave heating for the sugar extraction and the degradation of the partial starch in the peel. A catalyst was not required since the hydrolysis of starch, which breaks a bond in the molecule using water, is easier than that of cellulose since the starch can dissolve in water but the cellulose cannot. Moreover, the structure of cellulose is stronger than that of starch (Polymer Science Learning Center, 2005), and the dissolution of cellulose, therefore, necessarily requires a catalyst.

Generally, after thermal treatment, a lignocellulosic biomass may release sugars of various types, including glucose, fructose, xylose, galactose, arabinose, mannose and sucrose. However, the sugars that were detected in the liquid fraction using high performance liquid chromatography (HPLC), were of only three types, glucose, fructose and sucrose (total sugars), as determined by the Scientific Equipment Center, Prince of Songkla University. This may be due to the hemicellulose content from which xylose, galactose, arabinose and mannose are derived, not being hydrolyzed in the absence of an acid/enzyme.

This work investigated the effect of the variables in the microwave pretreatment on maximizing the sugars dissolved in the liquid fraction, and increasing the susceptibility of the peel to releasing soluble sugars after pretreatment. The liquid fraction, which might be wastewater in other works, was the desired product in this study, which can be used as the substrate for ethanol fermentation while the solid fraction with a high cellulose content can also be employed for ethanol production in future work.

3.2 Soluble sugar production

The responses in terms of reducing sugar and total sugar yields are summarized in Table 3. The results presented in Tables 1 and 3 show that water treatment using microwave heating promotes the release of sugars from the pineapple peel. A second-order model evaluating the effects of the variables and predicting the response was established from the experimental results as shown in Equation (2) for the reducing sugar yield, and Equation (3) for the total sugar yield. The coefficients and the regression analysis of the models, and the analysis of variance (ANOVA) are given in Tables 5 and 6, respectively. The accuracy of the model was estimated by the coefficient of determination (R^2) of the response and the overall fit of the model was also indicated by the results of the ANOVA. The significance of the individual, quadratic and interaction effects of the variables can be considered based on the probabilities (***P***-values) being less than 0.05 (Wang *et al.*, 2008).

Table 5. Regression analysis of the response surface models for the soluble sugar production using microwave-assisted water pretreatment.

Terms	Reducing sugar yield			Total sugar yield		
	Coefficient	Standard Error	***P***-value	Coefficient	Standard Error	***P***-value
Constant	-33.58000	8.29700	0.00488	281.09000	39.97000	0.00021
B	0.59700	0.10900	0.00094	-3.69000	0.52600	0.00021
W	-0.01639	0.00570	0.02375	0.09510	0.02744	0.01046
I	1.04000	0.30100	0.01059	4.46300	1.44900	0.01780
B^2	-0.00238	0.00040	0.00061	0.01133	0.00194	0.00065
W^2	0.00003	<0.00001	0.25700	-0.00007	0.00001	0.57300
I^2	-0.02967	0.00650	0.00260	-0.25000	0.03133	0.00094
BW	0.00011	0.00004	0.02457	-0.00047	0.00018	0.03285
BI	-0.00223	0.00187	0.27000	0.02268	0.00899	0.03964
WI	-0.00028	0.00015	0.10300	-0.00263	0.00071	0.00774
R^2	0.952			0.989		
R^2 adjusted	0.890			0.976		

Table 6. Analysis of variance (ANOVA) of the response surface models for the soluble sugar production using microwave-assisted water pretreatment.

ANOVA of the reducing sugar yield

Source	Degree of Freedom	Sum of Squares	Mean Square	***F***	***F****Signif*
Regression	9	25.37000	2.81900	15.34000	0.00080
Residual	7	1.28600	0.18400	-	-
LOF Error	5	1.28200	0.25600	130.02490	0.00765
Pure Error	2	0.00394	0.00197	-	-
Total	16	26.66000	-	-	-

ANOVA of the total sugar yield

Source	Degree of Freedom	Sum of Squares	Mean Square	***F***	***F****Signif*
Regression	9	2787.50000	309.72000	72.63000	0.00001
Residual	7	29.85000	4.26400	-	-
LOF Error	5	29.85000	5.96900	8221.89940	0.00012
Pure Error	2	0.00145	0.00073	-	-
Total	16	2817.30000	-	-	-

3.2.1 Response analysis of the reducing sugar yield

The model with good fit and a high determination coefficient ($R^2 = 0.952$) for the reducing sugar yield is represented by Equation (2):

$$\text{Reducing sugar yield} = -33.58000 + 0.59700B - 0.01639W + 1.04000I - 0.00238B^2 + 0.00003W^2 - 0.02967I^2 + 0.00011BW - 0.00223BI - 0.00028WI \quad (2)$$

where B, W and I are the biomass loading (g/L), microwave power (watts) and irradiation time (minutes), respectively.

The ***P***-values for the reducing sugar response (Table 5) showed that all effects of the variables had a significant influence on the reducing sugar yield except for the quadratic effects of microwave power and the interaction effect of the biomass loading and irradiation time. The interaction effects of the three variables are illustrated in Figure 2.

As shown in Figures 2(a) and 2(b), the optimal reducing sugar yields (>5%) were achieved with the biomass loading ranging from 120-140 g/L. This implies that the ratio of solids to water (or water amount providing H^+) influences the hydrolysis and the release of sugar by the peel, and that 1 L of water may not be sufficient for the treatment of peel in amounts higher than 140 g.

The reducing sugar yield decreased with increasing microwave power (Figures 2(a) and 2(c)). The power is the heating rate that controls the temperature and energy accumulation in the mixture. The operating times needed for the microwave unit used in this study to reach a steady state at a constant temperature of 97 °C are shown in Table 4. The time taken to reach a steady state depends on the operating volume and the thermal efficiency of the particular microwave unit used. An increase in the power may decrease the time (Table 4). Microwave irradiation directly accelerates the water molecules and their motion is converted suddenly into heat (Kappe, 2008), thus the treatment was able to operate efficiently at a lower power with a sufficient amount of water (biomass loading <140 g/L). However, a higher power was required for the treatment of biomass loadings with inadequate water (Figure 2 (a)). Moreover, the optimal yield (7-8%) was obtained at low power in the range of 90-270 watts for a time in the range of 8-15 minutes and a temperature of the mixture during treatment in the range of 35-55 °C as determined by an infrared thermometer (FLIR ix series IRC30) (Figure 2 (c)). The reason may be that temperature strongly influences solubility and the accumulation and degradation of sugar (Panpea *et al.*, 2008; Tian, Qi-hua, Ohsugi, Yamagishi, & Sasaki, 2006).

3.2.2 Response analysis of the total sugar yield

The experimental results as shown in Table 3 were used to establish the total sugar yield model, Equation (3), and its ANOVA results are shown in Table 6.

$$\text{Total sugar yield} = 281.09000 - 3.69000B + 0.09510W + 4.46300I + 0.01133B^2 - 0.00007W^2 - 0.25000I^2 - 0.00047BW + 0.02268BI - 0.00268WI \quad (3)$$

where B, W and I are the biomass loading (g/L), microwave power (watts) and irradiation time (minutes), respectively.

Form the ***P***-values (Table 5), the response of the total sugar yield is quite similar to the reducing sugar response with most effects being significant. There was however one insignificant effect which was the quadratic effect of the microwave power. The biomass loading, microwave power and time effects on the total sugar yield are illustrated in Figures 3(a)-3(c).

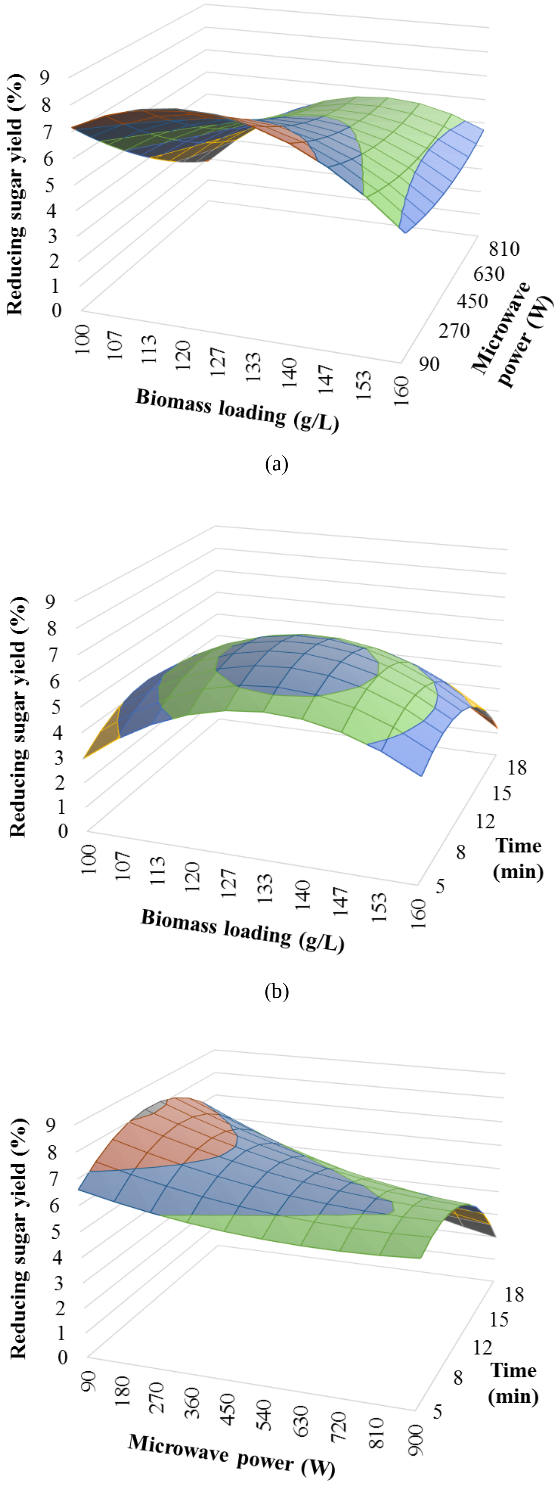

Figure 2. Reducing sugar yield in microwave-assisted water pretreatment as a function of; (a) biomass loading and microwave power for 13 min, (b) biomass loading and time with 450 W microwave power, and (c) microwave power and time using 130 g/L biomass loading.

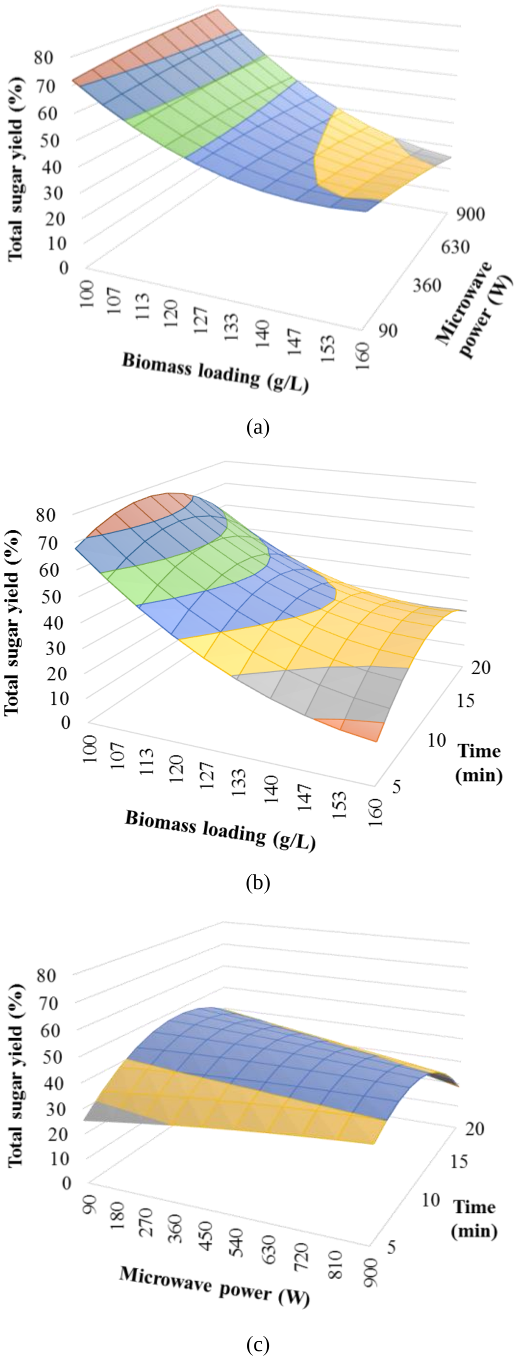

Figure 3. Total sugar yield in microwave-assisted water pretreatment as a function of; (a) biomass loading and microwave power for 13 min, (b) biomass loading and time with 450 W microwave power, and (c) microwave power and time using 130 g/L biomass loading.

As shown in Figures 3(a) and 3(b), the total sugar yield decreased markedly with increasing biomass loading. This result implies that the production of soluble total sugars requires the amount of water to exceed that of the reducing sugar. Higher soluble sugar levels were obtained with higher volumes of water, with the optimal total sugar yield (>70%) being achieved with a biomass loading below 107 g/L. The results were quite different from the reducing sugar response, probably because the effect of the power combined with the temperature had a lesser influence on the total sugar yield with its optimum being obtained at all powers (90-900 watts) for times in the range of 8-15 minutes (Figures 3(a) and 3(c)). The reason may be that the effect of temperature, which strongly influences the solubility and accumulation of total sugars, is different as between the three sugar types. Glucose, which is a monosaccharide, requires a lower temperature (<55 °C), while sucrose, which is a disaccharide, requires a higher temperature (>55 °C) (Panpea *et al.*, 2008). Consequently, the optimal yields were able to be obtained at various temperatures. This result suggests that varying the temperature of the mixture would offer alternative forms of the treatment designed to produce more mono- or di-saccharides as required.

3.2.3 Optimization of soluble sugar production

The best results based on the optimization of the models, produced the maximum reducing sugar yield of 8.3%, achieved using a biomass loading of 121.9 g/L at a microwave power of 90 watts for an irradiation time of 13 minutes. Meanwhile, the maximum total sugar yield of 80.2% was obtained using 100 g/L at 900 watts for 9 minutes. The two maximal responses were therefore acquired under quite different conditions. This is probably because the peel requires different temperatures for releasing glucose (which like fructose is a monosaccharide) and for releasing the total sugars, which also contain sucrose (a disaccharide). The mixture temperature rose to about 41 °C at a power of 90 watts for 13 minutes, whereas at 900 watts for 9 minutes the mixture temperature rose to the steady state temperature of 97 °C after 6 minutes and remained constant at 97 °C for the balance of the 9 minutes total time (Table 4).

However, since the sucrose can be hydrolyzed to glucose and fructose by *Saccharomyces cerevisiae* (common yeast) producing hydrolyzing enzymes such as sucrase, before fermentation to ethanol (Underkofler, Barton, & Rennert, 1958), the total sugar yield is the priority for soluble sugar production. Consequently, the optimal treatment was found to be 100 g/L at 900 watts for 9 minutes providing total sugar of 80.2%. Thus, 100 g of dried peel (575 g of raw pineapple peel) provided 80.2 g of total sugars in 1 L of liquid fraction consisting of 7.8 g glucose, 7.6 g fructose and 64.8 g sucrose. The optimum total sugars achieved of 13.9% w/w (13.9 g total sugars/100 g raw peel) was higher than that of 7.4% w/w achieved in previous work (Siti Roha, Zainal, Noriham, & Nadzirah, 2013).

The energy consumed in the optimal treatment to produce 1 g total sugars (fermentable sugars) was 6.0 kJ or 31.6 kJ to produce 1 g mono-sugars (sum of glucose and fructose), which was less than that recorded in prior work by Thangavelu *et al.* (2014) relating to the microwave hydrothermal hydrolysis of sago pith waste accelerated by carbon dioxide, which consumed 33 kJ per 1 g glucose. In addition, the energy consumption for the pineapple peel preparation (before the microwave treatment) to produce 1 g total sugars was 0.8 kJ for drying and 0.6 kJ for grinding. With these energy expenditures included, an estimate for the overall energy consumption in the soluble sugar production was about 7.4 kJ/g total sugars (or 39.1 kJ/g mono-sugars).

Moreover, the present work provides some advantages over previous works. Firstly, the microwave treatment was able to be limited to the boiling point of the sugar broth (the saturated temperature of the mixture) at a maximum temperature of 97 °C. This relatively low operating temperature can prevent the thermal decomposition of useful components in the biomass (Caddick, 1995). Further, since the treatment did not require a catalyst this can reduce costs, and does not generate unwanted by-products which inhibit fermentation, such as those which result from the use of acid catalysts (Da-Costa Sousa, Chundawat, Balan, & Dale, 2009). Additionally, with the condenser set fitted to the apparatus, the treatment can continue at a steady state without any loss of liquid, thus reducing energy consumption due to liquid evaporation, and also prevents corrosion from chemicals or vapor.

It is suggested that the initial total sugar concentration in the substrate for ethanol fermentation should be in a range between 125 and 250 g/L (Arroyo-López, Orlić, Querol, & Barrio, 2009) in order to obtain the optimal ethanol concentration for commercial production. The liquid fraction should, thus, be concentrated to increase the concentration of total sugars (>80.2 g/L) in the liquid substrate. Furthermore, the solid fraction remaining after the removal of the liquid fraction retains a high cellulose content even after the microwave treatment carried out in this study, and this can be additionally used as a feedstock to produce fermentable sugars for the production of ethanol in further work.

4. Conclusions

The results show that water pretreatment using simple microwave technology without the use of costly catalysts or expensive enzymes can be used to produce soluble sugars from pineapple peel being a biomass-waste. The liquid fraction produced in the pretreatment contained sufficient sugars and can be used as a substrate for ethanol fermentation. This offers an alternative cost-effective approach to the production of fermentable sugars with low energy consumption and a lower reagent requirement using a self-reliant technology.

Acknowledgements

This work was supported by the government budget of Prince of Songkla University, contract no. ENG570103S. The authors thank Mr. M. Currie and the Research and Development Office, Prince of Songkla University, for assistance with proof-reading the English.

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