

Novel hybrid process for the conversion of microcrystalline cellulose to value-added chemicals: part 1: process optimization

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Abstract In this paper, a novel hybrid process for the treatment of microcrystalline cellulose (MCC) under hot-compressed water was investigated by applying constant direct current on the reaction medium. Constant current range from 1A to 2A was applied through a cylindrical anode made of titanium to the reactor wall. Reactions were conducted using a specially designed batch reactor (450 mL) made of SUS 316 stainless steel for 30–120 min of reaction time at temperature range of 170–230 °C. As a proton donor H₂SO₄ was used at concentrations of 1–50 mM. Main hydrolysis products of MCC degradation in HCW were detected as glucose, fructose, levulinic acid, 5-HMF, and furfural. For the quantification of these products, High Performance Liquid Chromatography (HPLC) and Gas Chromatography with Mass Spectroscopy (GC–MS) were used. A ½ fractional factorial design with 2-level of four factors; reaction time, temperature, H₂SO₄ concentration and applied current with 3 center points were built and responses were statistically analyzed. Response surface methodology was used for process optimization and it was found that introduction of 1A current at 200 °C to the

reaction medium increased Total Organic Carbon (TOC) and cellulose conversions to 62 and 81 %, respectively. Moreover, application of current diminished the necessary reaction temperature and time to obtain high TOC and cellulose conversion values and hence decreased the energy required for cellulose hydrolysis to value added chemicals. Applied current had diverse effect on levulinic acid concentration (29.9 %) in the liquid product (230 °C, 120 min., 2 A, 50 mM H₂SO₄).

Keywords Biomass · Cellulose · Levulinic acid · 5-HMF · Sub-critical water · Hydrothermal electrolysis

Introduction

The growing interest in biomass as renewable feedstock for the production of value-added products results not only from environmental concerns but also from the need for political stability, which play an important role. With the uncertainties of petroleum-based economy, many nations have been transferring their petroleum-based economy to bio-based. Therefore, a nation lacking in of petroleum stocks can take advantage of its natural sources (biomass) to produce such value-added products as fermentable sugars and chemicals. In this manner, conversion of different kinds of biomass sources to high value-added chemicals has been investigated in literature for last decades

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(Wu et al. 2010; Yan et al. 2010). So far, mineral acids (Muranaka et al. 2014), solid acid catalysts (Liu et al. 2015; Zi et al. 2015), enzymes (Annamalai et al. 2014; Badgujar and Bhanage 2016), subcritical water (Saito et al. 2009) have been used to hydrolyze biomass into value-added chemicals such as Levulinic acid, Hydroxymethylfurfural (5-HMF), Furfural.

Sub- and super-critical water have gotten much attention due to the versatile properties such as dielectric constant ($\epsilon \cong 10$ near critical) and the ion product concentration which is 10^{-11} in the temperature range between 200 and 300 °C (Marshall and Franck 1981). Increase in temperature leads to decrease in pH value of water by three units, providing more H_3^+ ions for acid catalyzed reactions. Hence, the ionic product concentration can be used for optimizing acid–base reactions (Kruse and Dinjus 2007). In addition, the current efficiency increases at near critical conditions that result in more economically feasible electrochemical reactions (Asghari and Yoshida 2008). Due to these versatile properties of water at near critical region, electrolysis reaction of microcrystalline cellulose was conducted under hydrothermal conditions.

Treatments of cellulose in both hot compressed water and supercritical water have shown to be an efficient and environmentally friendly way to utilize cellulose for value-added chemicals (Kumar et al. 2011; Reddy et al. 2014). Reddy et al. (2014) showed that hydrolysis of cellulose at supercritical region favors the radical based reactions in which gaseous products as hydrogen and carbon dioxide were produced. Yin and Tan (2012) investigated cellulose hydrolysis at neutral, acidic, and alkaline mediums under hydrothermal conditions and found that reaction pathway changed in favor of 5-HMF and levulinic acid under acidic environment. However, 5-HMF polymerized to tarry materials at higher reaction temperature and longer time. Under alkaline condition, reaction favored the formation of carboxyl acids such as lactic and acetic acids. However, bio-oil yield could not exceed 35 % under both acidic and alkaline environment. Therefore, parameters as reaction temperature, time and pH should not be separately investigated. Mante and Agblevor (2011) investigated the interaction parameters in pyrolysis of wood shavings and showed that liquid product yield is mainly influenced by temperature, feed rate and gas flow rate. Thus, in this study, hydrothermal electrolysis of cellulose was investigated by using factorial

design to reveal interaction parameters between applied current, reaction temperature, time and acid concentration.

Electrochemical methods that are used for the oxidation and synthesis of organic compounds (Sasaki et al. 2007) have become attractive due to promising features that overcome the problems associated with conventional methods such as corrosion, long reaction times, and emission of greenhouse gases. As a hybrid technology, hot compressed water properties such as high ionic product concentration supply a high efficient medium in terms of migration of electro-active species. Thus, high current efficiency in electrochemical processes minimizes the amount of energy consumed for electrochemical reactions (Asghari and Yoshida 2008).

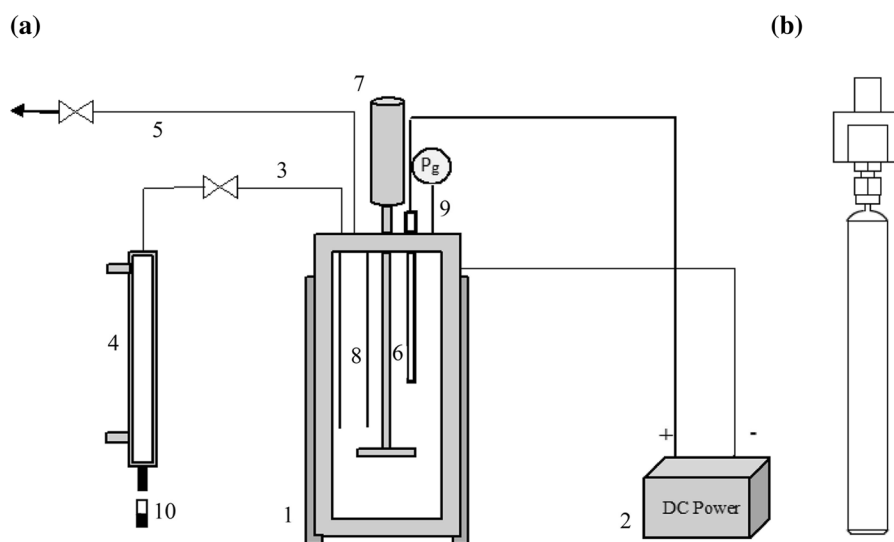
Studies on hydrothermal electrolysis of organic compounds (Sasaki et al. 2010; Yuksel et al. 2010) showed a promising method for the production of value added chemicals in terms of increased selectivity. Yuksel et al. (2010) studied electrolysis of glycerol in sub-critical water and reported that application of 1 A current to reaction medium increased the yield of lactic acid by 9 % compared to experiments without current. Sasaki and coworkers (Sasaki et al. 2007) studied hydrothermal electrolysis of glucose and found that degradation products such as organic acids were produced with a yield of 40–60 %.

In this study, hydrothermal electrolysis of microcrystalline cellulose to value-added chemicals was investigated in detail and it was aimed to understand the effect of applied current on the product distribution, cellulose and TOC conversions. In this manner, product yields were statistically analyzed via Analysis of Variance (ANOVA) by considering interaction of applied current (0–2A) with other parameters such as H_2SO_4 concentration (1–50 mM), reaction temperature (170–230 °C) and time (30–120 min.). Response surface methodology was used to optimize process parameters in a way of maximizing valuable products as levulinic acid, 5-HMF, and furfural.

Materials and methods

Microcrystalline cellulose (MCC, Sigma Aldrich) was used in powder form (particle size $<20 \mu m$). Sulfuric Acid (Merck, 96–98 % purity) was used as eluent in HPLC (Agilent 1200). Analytical grade ($>99 \%$,

Fig. 1 **a** Hydrothermal electrolysis reactor: (1) electrical heater, (2) DC Power controller, (3) liquid sample tubing, (4) heat exchanger (5) gas sample collector, (6) anode, (7) rotor, (8) thermocouple, (9) pressure gauge, (10) sample holder, **b** Specially designed cylindrical electrode (anode)



(Sigma Aldrich) of standard chemicals as glucose, fructose, 5-HMF, levulinic acid, furfural, were used for the preparation of reference standards to build calibration curves for HPLC analysis to quantify the product yields.

In HPLC analysis, 3.75 mM H_2SO_4 with the flow rate of 0.5 ml/min was used as eluent. Sugar column (Shodex, SH1100) with column temperature of 50 °C was selected for clear separation of products. Refractive index (RID) was used for detection of aldehydes and organic acids. GC–MS (Agilent 6890 N/5973 N Network) was used to define the products that were unidentified in HPLC analyses. Gas products were analyzed by Gas Chromatography equipped with a Thermal Conductivity Detector (GC-TCD) (Agilent 6890 N). TOC analyzer (Shimadzu TOC- VCPH) was used to monitor TOC conversions in the liquid products and solid residues. Morphological properties of solid residues were analyzed by Scanning Electron Microscopy (SEM), (Quanta 250 SEM) with ETD detector. The chemical composition of solid residues obtained at the end of the reaction was analyzed by Fourier Transmission Infrared Spectroscopy (FT-IR). FT-IR analyses were conducted with scan rate of 4 cm^{-1} from $600\text{--}4000\text{ cm}^{-1}$ wavenumber by Perkin Elmer Frontier-Spectra two.

Experimental procedure and equipment

Hydrothermal electrolysis experiments were carried out in a 450 mL of batch reactor (Parr 5500 series)

made of SUS 316 stainless steel, which is illustrated in Fig. 1. All experiments were conducted in 200 ml of deionized water containing 8 gr of cellulose. Sulfuric acid was added to the reaction medium as external proton source at different concentrations (1–50 mM).

Reaction medium was heated to desired temperature with an external electrical heater and reaction time was set as zero when the desired reaction temperature is reached. In order to observe the effect of heating period, first sample was taken at the time of desired temperature is reached. 2 ml of samples were collected at reaction times of 0, 15, 30, 60, 90, and 120 min.

Hydrothermal electrolysis experiments were carried out at a constant current (0–2 A) passing through the electrodes. Specially designed cylindrical type electrode (12 mm diameter, 94 mm length), made of titanium, was used as anode and cylindrical reactor wall (165 mm in length, 76 mm outer diameter) was acted as cathode.

Experimental design

In this study, effect of parameters as reaction Current (A), Temperature (B), Time (C) and H_2SO_4 concentration (D) on responses such as conversion of cellulose, conversion of TOC, yields of 5-HMF, levulinic acid and furfural were statistically analyzed by using surface response methodology (RSM) to optimize desired responses. Experimental design was built by $1/2$ fractional factorials design with two level of

Table 1 Experimental design and responses as percent yield of liquid products

Design	Temperature (°C)	Time (min.)	H ₂ SO ₄ (mM)	Current (A)	Conversion	TOC	Glucose	Fructose	5-HMF	Furfural	Levulinic acid
–	170	30	1	0	11.38	29.00	0.36	0.48	0.04	0.07	0.04
+++	230	30	1	2	81.25	60.71	12.20	7.94	7.00	1.51	3.00
-++	170	120	1	2	11.25	8.10	1.71	1.94	0.18	0.16	0.68
++-	230	120	1	0	70.63	40.48	8.47	*	5.29	2.55	11.50
-++	170	30	50	2	29.00	22.00	4.17	3.21	0.74	0.35	1.57
++-	230	30	50	0	77.13	61.50	4.64	1.74	3.57	2.20	36.00
-++	170	120	50	0	48.75	29.07	6.36	0.95	3.57	0.92	7.20
++++	230	120	50	2	74.38	39.38	0.60	0.24	0.27	0.48	28.98
—+	170	30	1	0	11.38	28.81	0.20	0.28	0.03	0.07	0.04
+++	230	30	1	2	78.75	61.20	11.93	6.03	7.44	1.48	3.20
0++	170	120	1	2	7.50	8.21	1.58	1.90	0.17	0.13	0.50
++-	230	120	1	0	68.75	41.00	9.00	*	5.50	2.45	12.00
-++	170	30	50	2	30.75	22.35	4.20	3.69	0.96	0.48	1.40
++-	230	30	50	0	82.63	62.02	4.63	2.85	2.82	2.43	37.00
-++	170	120	50	0	46.88	28.48	6.60	1.90	3.63	0.83	7.10
++++	230	120	50	2	77.00	39.52	0.44	0.26	0.35	0.47	29.90
0000	200	75	25.5	1	81.13	62.62	15.41	7.79	3.71	1.05	12.44
0000	200	75	25.5	1	81.38	62.20	15.70	7.20	3.50	1.04	*
0000	200	75	25.5	1	80.00	62.00	15.20	7.50	4.00	0.92	11.91

* Data does not exist

(+) High, (0) Middle, (-) Low values of design variables

four factors (A, B, C, and D) with three center points and two replicates for corner points. A total number of 19 experimental runs were built and given in Table 1.

Results and discussion

HPLC and GC–MS analysis

Liquid products of cellulose decomposition contain variety of hydrocarbons. In this study, the key components of the reaction pathway such as glucose, fructose, levulinic acid, 5-HMF, and furfural were

identified and quantified via HPLC (Fig. 2) and GC–MS analyses. HPLC results were verified by GC–MS results of products point of interest (Table 2). In order to increase the sensitivity of GC–MS, Selected Ion Methodology (SIM) was used for the quantification of products.

Yield of product was calculated based on the carbon amount of species produced and initial amount of carbon in cellulose (Eq. 1). Carbon amount of cellulose was found as 42–45 % by TOC analysis. For all experiments, carbon balance was calculated as above 90 %. Conversion of cellulose was calculated by using Eq. 2.

$$\text{Yield of Product \%} = \frac{(\text{Number of carbon of species}) * (\text{Mole of species produced})}{\text{Moles of carbon in cellulose}} * 100 \quad (1)$$

$$\text{Converted Cellulose \%} = \frac{\text{Initial amount of cellulose (gram)} - \text{Residual amount at the end (gram)}}{\text{Initial amount of cellulose (gram)}} * 100 \quad (2)$$

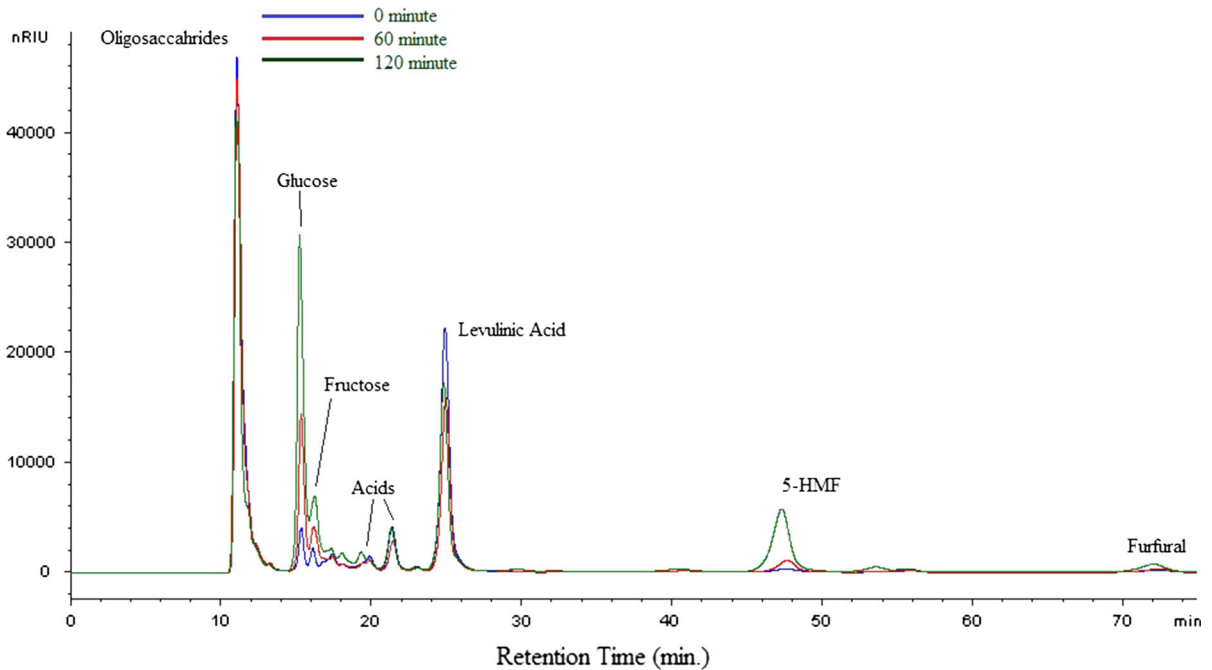


Fig. 2 HPLC chromatogram of cellulose decomposition products at 170 °C, with 50 mM H₂SO₄ at sampling time of 0, 60 and 120 min

Table 2 GC–MS–SIM data of hydrolysis products of cellulose

Species	Molecule formula	Retention time (min.)
Glyceraldehyde	C ₃ H ₆ O ₃	13.07
Glycoaldehyde	C ₂ H ₄ O ₃	13.07
Lactic acid	C ₃ H ₆ O ₃	29.72
Glycerol	C ₃ H ₈ O ₃	31.29
Formic acid	CH ₂ O ₂	16.89
Acetic acid	C ₂ H ₄ O ₂	15.1
Levulinic acid	C ₅ H ₈ O ₃	31.62
5-HMF	C ₆ H ₆ O ₃	36.09
Furfural	C ₅ H ₄ O ₂	15.53

Analyses of variance (ANOVA)

Cellulose conversion

Table 3 indicates summary of ANOVA test for cellulose conversion values. On this table,

p-values (less than $\alpha = 0.05$) of each affecting parameters are given individually to consider the interactions between the parameters. The R² with a value of 0.99 indicates that model is well fit to the observed response. The model accuracy is also confirmed by histograms including normal probability and residual plots.

Table 3 Statistical analysis results for converted cellulose with 95 % of confidence level

Source	DF	Sum of squares	Mean of squares	F value	P value
Model	8.00	96.71	12.09	542.06	0.000
Linear	4.00	75.06	18.76	841.37	0.000
Current	1.00	0.31	0.31	13.69	0.004
Temperature	1.00	68.43	68.43	3068.57	0.000
Time	1.00	0.00	0.00	0.15	0.708
H ₂ SO ₄	1.00	6.31	6.31	283.06	0.000
Square	1.00	14.92	14.92	668.79	0.000
Current × current	1.00	14.92	14.92	668.79	0.000
2-way interaction	3.00	6.74	2.25	100.75	0.000
Current × Temperature	1.00	1.09	1.09	48.73	0.000
Current × Time	1.00	4.17	4.17	187.06	0.000
Current × H ₂ SO ₄	1.00	1.48	1.48	66.47	0.000
Error	10.00	0.22	0.02		
Total	18.00	96.93			

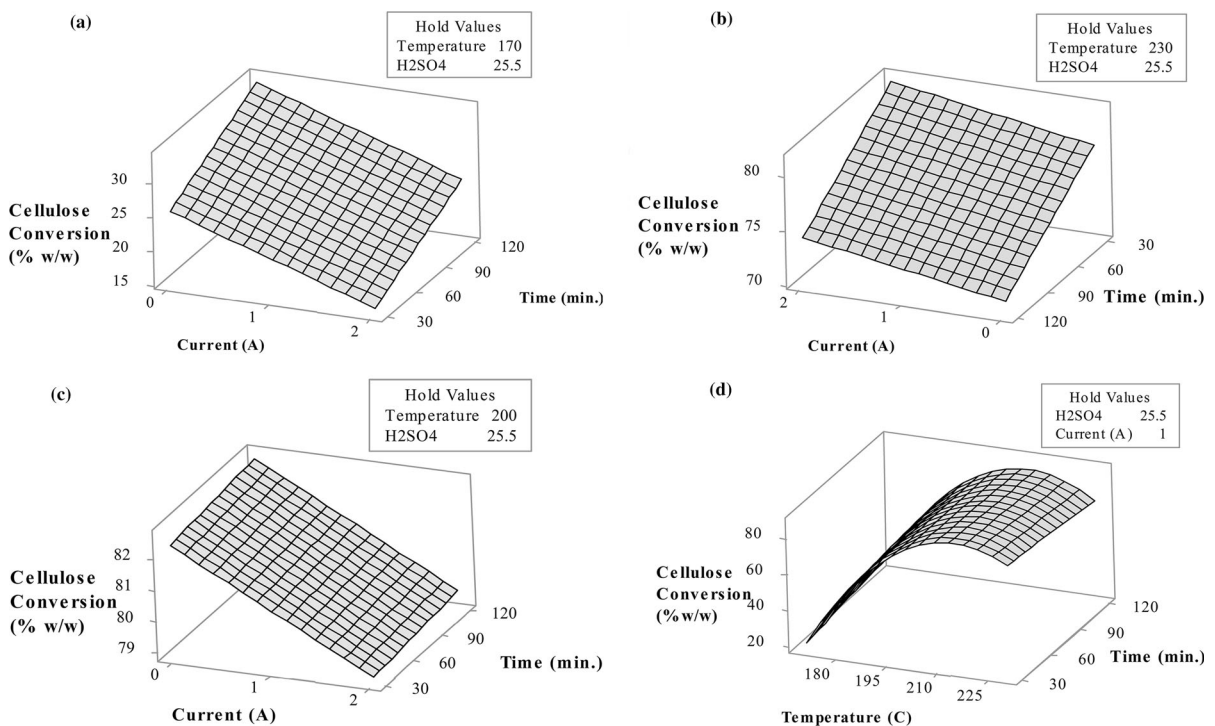


Fig. 3 Surface response plot of cellulose conversion with the hold values of 25 mM H₂SO₄ at **a** 170 °C, **b** 230 °C, **c** 200 °C, and with **d** 1 A current

T-tests of operating parameters (current, temperature, H₂SO₄, time) defined the coefficient of main effects and their second orders with interaction terms in

the model given as Eq. 3. The predicted cellulose conversion values can be found by using proposed model equation (Eq. 3) in the range of fitted boundaries.

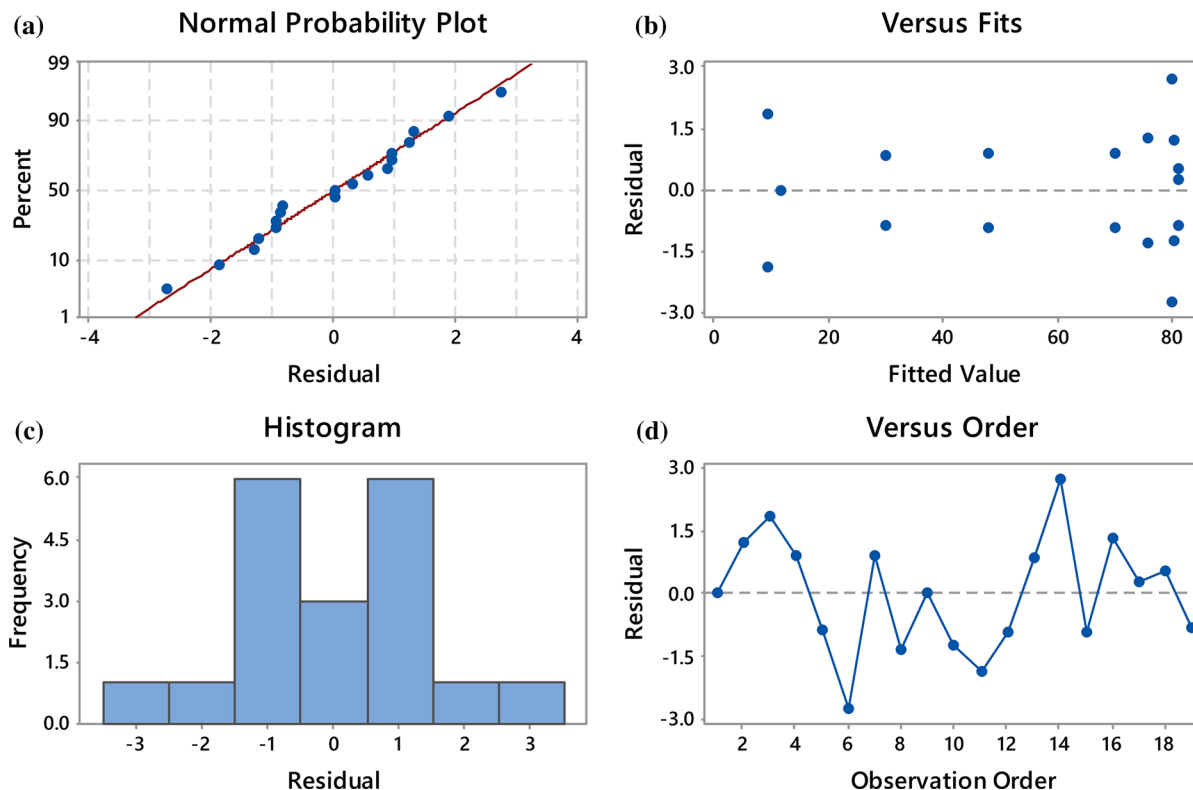


Fig. 4 Histogram plots of converted cellulose, **a** normal probability plot of residuals, **b** residuals versus predicted values, **c** frequency histogram of residuals, **d** observation orders of residuals

$$\begin{aligned}
 \text{Converted Cellulose} = & -9.721 + 4.152A + 0.06025B \\
 & + 0.01167C + 0.03806D \\
 & - 2.4298A^2 + 0.00869A * B \\
 & - 0.011347A * C \\
 & - 0.01242A * D
 \end{aligned}
 \tag{3}$$

As seen in Table 3, in the model, some of the interaction terms were removed due to the lack of fit ($p > 0.05$) and considered as disturbance in the system causing error in the measured responses. As an individual parameter, reaction time has the p value of 0.708, which is higher than the confidence interval indicating that time has no significant effect on the conversion of cellulose. However, interaction parameter (current*time) showed that coupled effect of current and time was significant and thus, model did not avoid time as insignificant parameter. In order to visualize the effect of parameters, response surface plots (Fig. 1) of cellulose conversion was plotted by

constant concentration of 25 mM H_2SO_4 at different reaction temperatures of 170, 200 and 230 °C. Temperature had the highest impact on cellulose degradation since temperature increases up to 220 °C as ionic product concentration increases in sub-critical water region (Dinjus and Kruse 2004). In order to depolymerize MCC, hydronium ions (H_3O^+) must access to inter- or intra-molecular hydrogen bonding. Thus, diffusion of protons (H^+) plays a crucial role together with versatile properties (low density, high diffusivity) of water in sub-critical region.

Increase in the current might have increased H^+ ion concentration due to the increase in potential difference, which resulted in the formation of acid layer around the anode material (Mabbott 1983). The diverse effect of current was more observable at 170 °C (Fig. 1a) in comparison to 200 °C (Fig. 1c) since H^+ concentration was lower at low temperature of reaction. On the contrary, current had positive effect on cellulose conversion at 230 °C (Fig. 1b). Current might have restricted the formation of the char as a

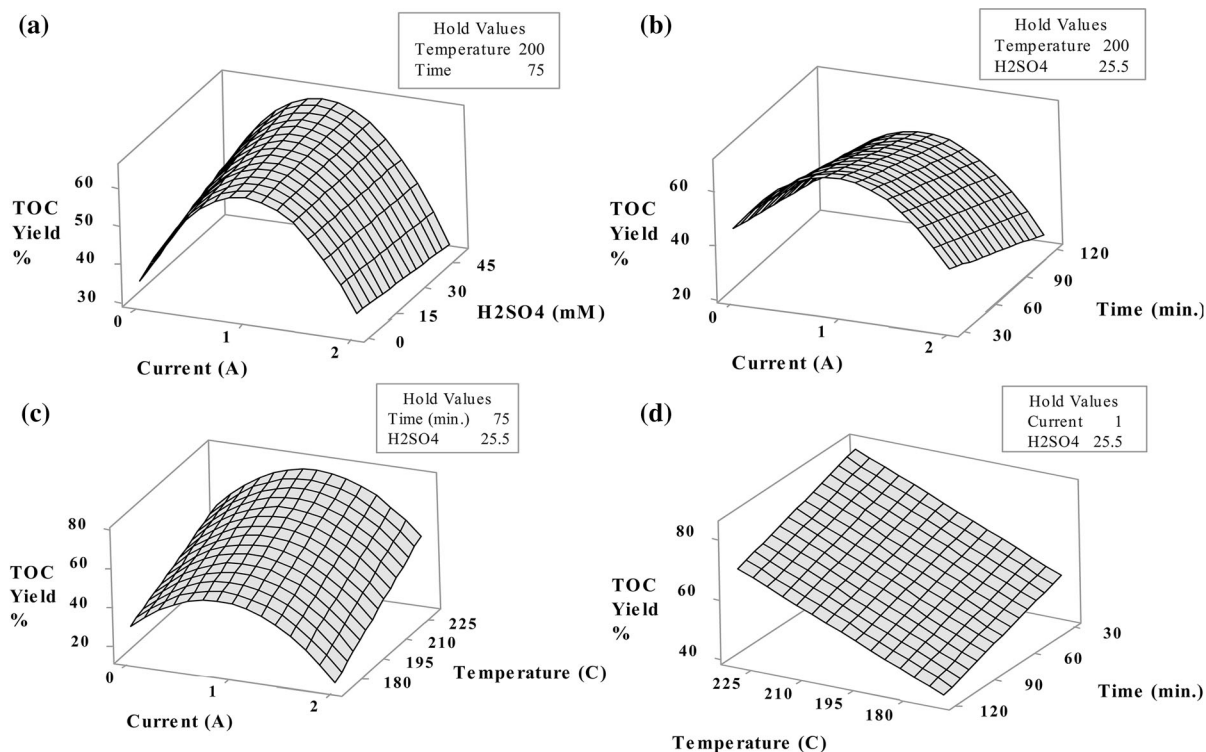


Fig. 5 Surface response plots of total organic content (TOC) with effecting parameters of applied current, reaction temperature and time, acid concentration (H₂SO₄)

result of the polymerization of 5-HMF at higher reaction temperatures (Chuntanapum et al. 2011). Moreover, Fig. 1d indicates that conversion reached its highest value at 200 °C and decreased at higher reaction temperatures due to formation of char. Additionally, for longer reaction time, conversion started to decrease due to further formation of char molecules (Fig. 1d). Although the mechanism has not been revealed yet, it is possible to say that applied current on the reaction medium at higher temperatures can alter the formation of char (Fig. 3). Moreover, effect of current on char formation may be related to 5-HMF concentration behavior under applied current, which will be discussed in detail in “5-HMF, levulinic acid, furfural production” section

TOC conversion, glucose and fructose yields in liquid product

ANOVA test of ($p > 0.05$) TOC conversion showed that the most significant term was temperature followed by reaction time and current applied (Online

resource). Quadratic model equation was built with the R^2 value of 0.99 and Eq. 4 was evaluated as model equation. The histogram plots of model were analyzed showing that residuals were normally distributed. This indicates that model as applicable, as seen Fig. 4.

$$\begin{aligned}
 \text{TOC Conversion} = & -31.280 + 33.774A + 0.37351B \\
 & - 0.11753C + 0.21319D \\
 & - 25.908A * A + 0.10524A \\
 & * B - 0.03992A * C - 0.14478A * D
 \end{aligned}
 \quad (4)$$

Surface response plot (Fig. 5c) of TOC formation indicates that as the temperature increased, TOC conversion increased as well due to the fact that protonation of $\beta(1-4)$ Glycosidic bonds resulted in the formation of decomposition products. However, increase in reaction time had diverse effect on TOC conversion that could be attributed to the formation of char and gas products at longer reaction times during decomposition of cellulose in hot compressed water conditions (Wiedner et al. 2013). Application of current to the reaction medium dramatically

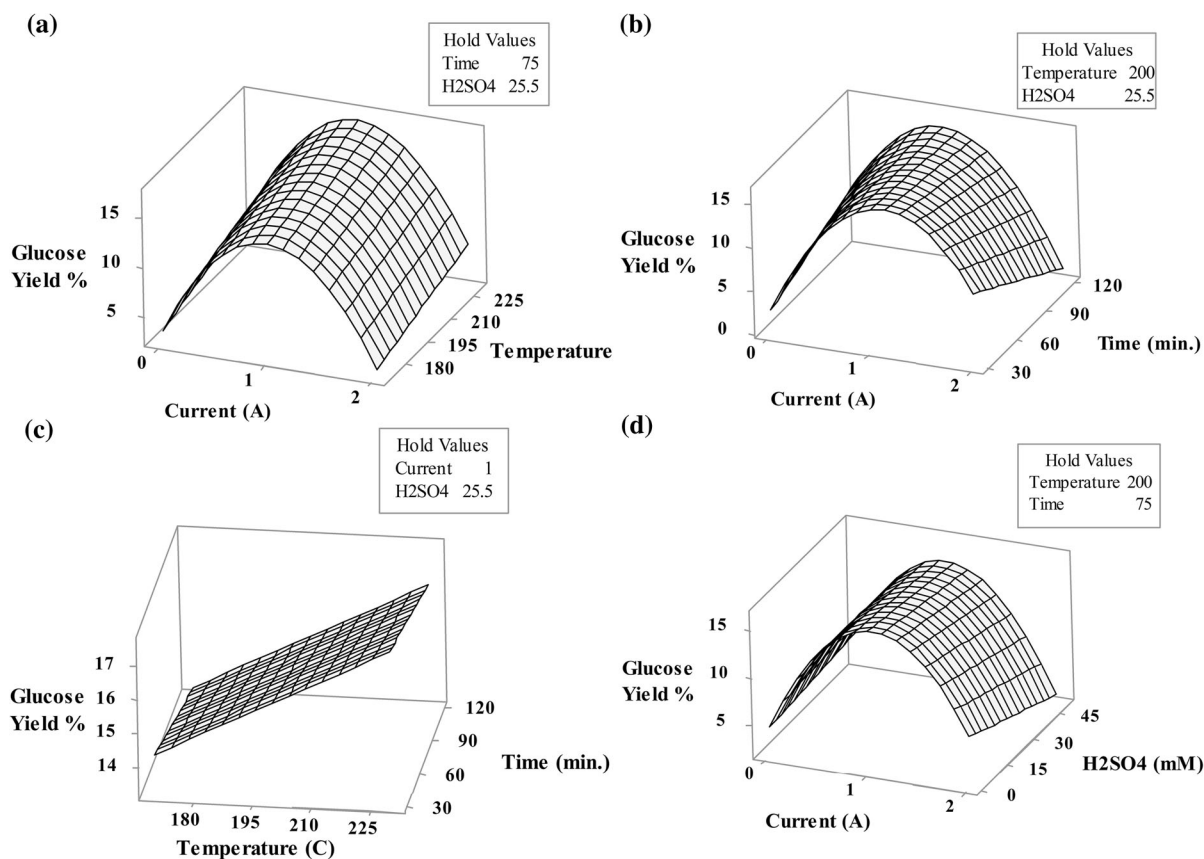


Fig. 6 Surface response plots of glucose yield with effecting parameters of applied current, reaction temperature, time and acid concentration (H_2SO_4)

increased the TOC conversion (Fig. 5a) up to 1 A. However, further increment in current resulted in sharp decrease of TOC conversion. Similar trend was also observed for all reaction temperatures (Fig. 5c). The current effect on TOC conversion at low acid concentration of 1 mM H_2SO_4 supported the idea of electrochemically generated acid layer around anode material due to dissociation of water molecules that resulting in the protonation of $\beta(1-4)$ Glycosidic bonds. It was also found that conversion of cellulose was diversely affected by the applied current, which can be attributed to the increase in TOC conversion selectively with direct current passing through the electrodes by limiting the formation of tarry material. 5-HMF formation and SEM images (“[Analysis of solid residue](#)” section, Fig. 12c) further supported the formation of tarry material for non-current experiments.

In electrochemical organic synthesis reactions, water as a solvent has a poor potential window; in other words, applied over potential results in the formation of hydrogen and oxygen gases. High

potential difference in reaction medium may increase the formation rate of hydrogen gases and hence, protonation reaction rate increases due to the high concentration of H^+ ions in reaction medium. Therefore, diverse effect on TOC conversion could be due to further reactions involved as a result of high H^+ concentration at longer reaction time (75 min.). Shorter reaction time (30 min) with passing 2 A current resulted in TOC conversion of 61 % with 1 mM H_2SO_4 and at a temperature of 230 °C. On the other hand, the current free experiment at the same conditions except 50 mM H_2SO_4 concentration yielded 62 % of TOC conversion (Table 1). Applied current diminished the acid concentration from 50 to 1 mM by keeping the TOC amount constant. It is the evidence of the over potential resulting in the formation of acid layer around anode. The use of model equation for TOC prediction (Eq. 4) by applying 2 A current at 230 °C with 1 mM H_2SO_4 resulted in 64 % of TOC at reaction time of 10 min while 1 A current

increased to 84 %. This might be explained by acid formation due to dissociation of water around the anode and increase in the rate of formation of hydrogen gases at higher current values as observed in GC-TCD analysis. Moreover, GC-TCD analysis showed that there was no hydrogen gas formed during 1 A applied current in contrast to 2 A in which hydrogen gases was formed with a concentration of 95 µg/ml (“Gas products” section, Fig. 11a.)

As well known, cellulose decomposes to the cellobiose and oligosaccharides such as celotriose, cellopentose as a result of protonation of intra- and inter-molecular hydrogen bonds. Further decomposition resulted in formation of glucose molecules. ANOVA test to glucose yield ($p > 0.005$) showed that all parameters had significant effect as individually; however, interaction term of current with temperature had the p value of 0.713 that was higher than confidence interval. Therefore, it was manually removed from the model and its effect was considered as disturbance in responses and investigated model equation (Eq. 5) was given as follows.

$$\begin{aligned} \text{Glucose Yield\%} = & -10.943 + 27.548A + 0.05569B \\ & + 0.05722C + 0.02143D \\ & - 10.619A * A - 0.06774A * C \\ & - 0.05666A * D \end{aligned} \quad (5)$$

ANOVA results showed that temperature had the highest effect on glucose yield [Online Resource (Table 5)]. Response surface plot of glucose (Fig. 6c) indicates that at low concentration of acid (1 mM H₂SO₄), when reaction temperature was increased, glucose concentration also increased from 4 to 9 %. At higher reaction temperature, up to 230 °C, dissociation constant, K_w , for water increases (Bignold et al. 1971). Formation of water dissociation products as H⁺ and OH⁻ favor the cellulose decomposition reaction pathway in ionic reaction mechanism (Buhler et al. 2002). Therefore, hydrolysis rate of cellulose increases at higher temperature and yields high concentration of glucose. In acidic conditions, glucose is further involved in the reactions such as isomerization to fructose following the formation of decomposition products such as 5-HMF, levulinic acid, formic acid, and furfural. Hence, glucose yield decreased to 2 % at higher acid concentrations (25 and 50 mM H₂SO₄). As the ionic reaction mechanism

becomes favorable at higher K_w value of water, applied current to the reaction medium may favor the decomposition pathway in ionic mechanism due to the self-dissociation of water around the anode and cathode. In addition, current had similar effect on glucose yield (Fig. 6a, b, d) as it was in TOC yield. Applied 1 A current on the reaction medium at low temperature (170 °C) increased the glucose yield (13 %) compared to current free experiment (3.2 %). Electrochemical generation of acid layer around anode material could have increased the rate of cellulose decomposition as similar current effect was observed on TOC conversion (Fig. 5a). However, 2 A of current loaded experiments for longer reaction times (75 and 120 min.) resulted in lower glucose yield. This could be due to further reactions of glucose such as retro-aldol condensation to erythrose, glycoaldehyde, and glyceraldehyde (Yin and Tan 2012).

Fructose is produced by isomerization of glucose. Further decomposition and dehydration of fructose resulted in products as furans and carboxylic acids (Promdej and Matsumura 2011). The Lobry de Bruyn-Alberda van Ekensteing (LBAE) transformation of glucose to fructose has been studied by number of researches and found that intermediate products of enediols are produced during this transformation. Retro aldol and reverse aldol condensation of aldehydes yield erythrose, glycoaldehyde and glyceraldehyde (Kabyemela et al. 1999). GC-MS results showed that glycoaldehyde and glyceraldehyde were formed during hydrothermal electrolysis of microcrystalline cellulose. In ionic mechanism, retro aldol condensation of enediols, which is formed during LBAE transformation, yields to the formation of aldehydes. In addition, ANOVA test to fructose yield ($p > 0.05$) showed that temperature and acid concentration were the most significant terms in the model equation (Eq. 6). Fructose formation was effected significantly since the ionic concentration highly depends on the temperature and acid concentration. However, the individual effect of current and reaction time had the p -value of 0.982 and 0.579, respectively. Therefore current and reaction time had no significant individual effect on the fructose yield. This is due the fact that, an increase in the applied current at low temperature (170 °C) had no significant effect on fructose formation at different reaction times. This could be explained by the lack of reactant such as glucose for the isomerization reaction. Therefore, ANOVA

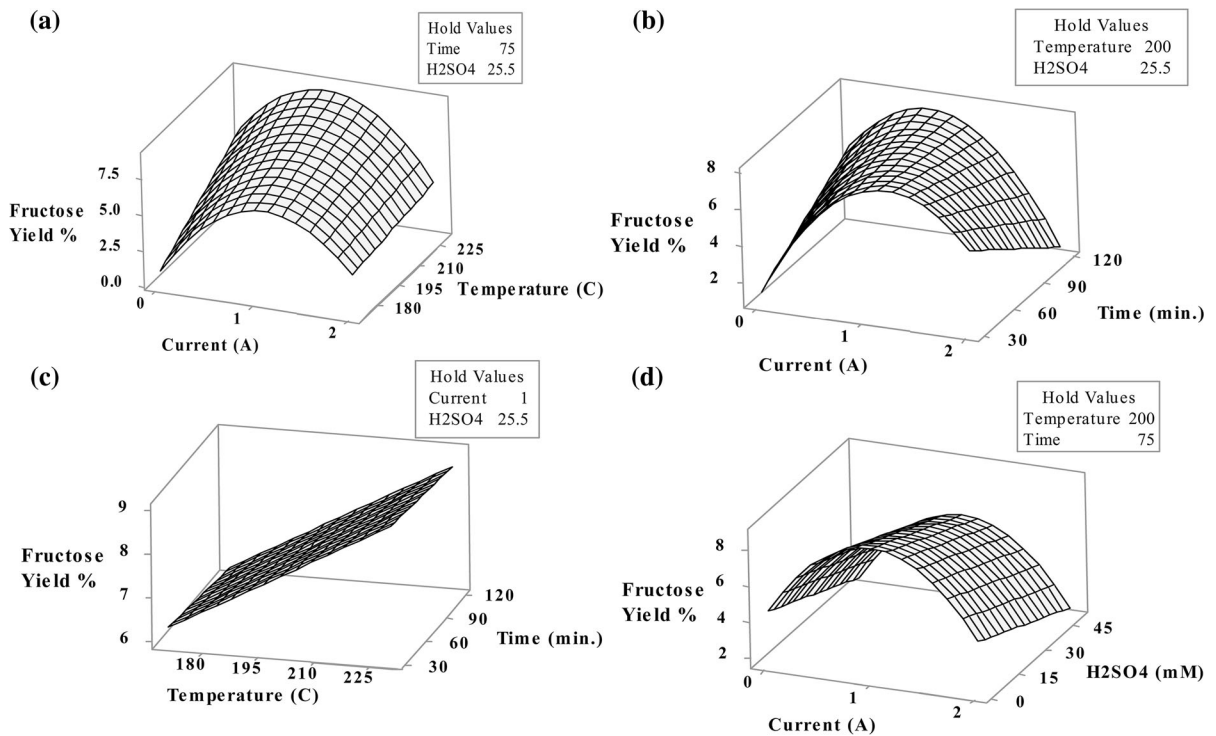


Fig. 7 Surface response plots of fructose yield with effecting parameters of applied current, reaction temperature and time, acid concentration (H₂SO₄)

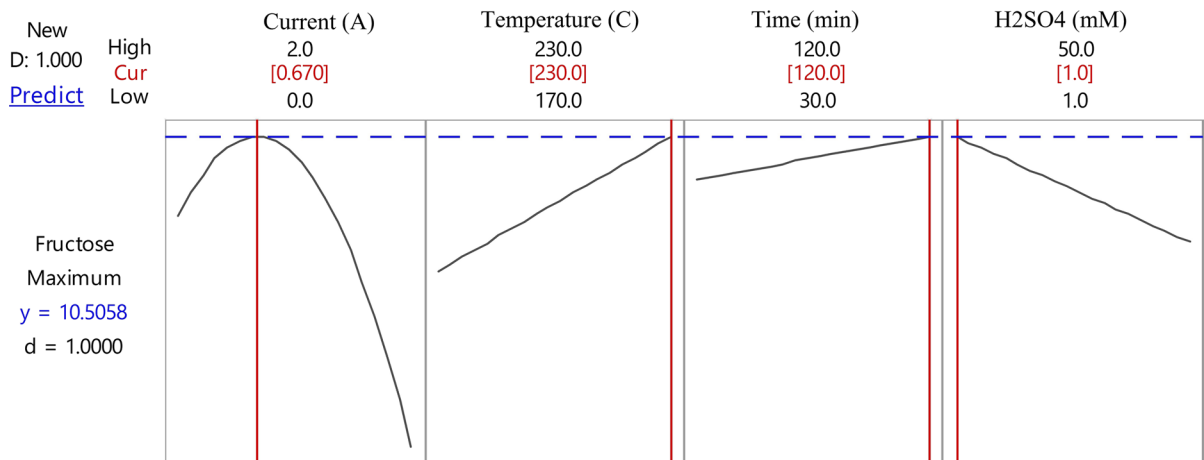


Fig. 8 Predicted operating parameters to maximize fructose yield

showed that current and reaction time had no significant individual effect on fructose formation; however, coupled effect of current and reaction time with reaction temperature and acid concentration resulted in the p-value less than 0.05 indicating that synergetic effect plays an important role in glucose isomerization

reaction (Online Resource). As a result, applied current became significant at higher reaction temperatures (200 and 230 °C). At 200 °C and 1 A of current, glucose concentration reached its maximum value and was isomerized to fructose due to the higher concentration of reactant.

Table 4 Statistical analysis results of 5-HMF, levulinic acid and furfural

Source	(5-HMF)					Levulinic acid				
	DF	Sum of squares	Mean of squares	F value	P value	DF	Sum of squares	Mean of squares	F value	P value
Model	8	105.83	13.23	238.16	0.000	8	2774.65	346.83	2508.62	0.000
Linear	4	45.96	10.74	193.34	0.000	4	2275.66	568.92	4114.95	0.000
Current (A)	1	3.37	3.37	60.62	0.000	1	108.42	108.42	784.20	0.000
Temperature (B)	1	32.83	32.83	591.09	0.000	1	1278.96	1278.96	9250.67	0.000
Time (C)	1	0.83	0.83	14.91	0.003	1	15.23	15.23	110.15	0.000
H ₂ SO ₄ (D)	1	5.93	5.93	106.74	0.000	1	873.05	873.05	6314.79	0.000
Square	1	3.28	3.28	59.02	0.000	1	1.50	1.50	10.84	0.000
Current × current	1	3.28	3.28	59.02	0.000	1	1.50	1.50	10.84	0.009
2-Way interaction	3	59.60	19.87	357.63	0.000	3	497.49	165.83	1199.45	0.000
Current × temperature	1	0.60	0.60	10.81	0.008	1	28.06	28.06	202.98	0.008
Current × time	1	44.55	44.56	802.31	0.000	1	464.08	464.08	3356.68	0.000
Current × H ₂ SO ₄	1	14.44	14.44	259.96	0.000	1	5.35	5.35	38.68	0.000
Error	10	0.56	0.06			9	1.24	0.14		0.000
Total	18	10.39				17	2775.89			

Source	Furfural				
	DF	Sum of squares	Mean of squares	F value	P value
Model	8	12.89	1.61	290.99	0.000
Linear	4	9.60	2.40	433.68	0.000
Current (A)	1	2.61	2.61	471.08	0.000
Temperature (B)	1	6.97	6.97	1258.81	0.000
Time (C)	1	0.02	0.02	4.06	0.000
H ₂ SO ₄ (D)	1	0.00	0.00	0.76	0.000
Square	1	0.00	0.00	0.49	0.000
Current × current	1	0.00	0.00	0.49	0.498
2-Way interaction	3	3.28	1.09	197.58	0.498
Current × temperature	1	1.51	1.51	273.25	0.000
Current × time	1	1.30	1.30	234.73	0.000
Current × H ₂ SO ₄	1	0.47	0.47	64.75	0.000
Error	10	0.06	0.02	0.01	0.000
Total	18	12.94	0.055		

$$\begin{aligned}
 \text{Fructose Yield\%} = & -13.58 + 17.89A + 0.0753BA \\
 & + 0.04053C - 0.05311D \\
 & - 4.340A * A - 0.02987A \\
 & * B - 0.04322A * C
 \end{aligned}
 \tag{6}$$

The interaction of current and acid concentration had no significant impact ($p > 0.05$) and therefore removed

from the model equation. Response surface plot (Fig. 7a) indicates that current had high interaction with reaction temperature and time (Fig. 7b). When the process conditions were optimized to maximize fructose yield, Fig. 8 was obtained. The model resulted in 10 % of maximum fructose yield under the conditions of 0.67 A current, reaction temperature as 230 °C, for 120 min reaction time and with 1 mM H₂SO₄.

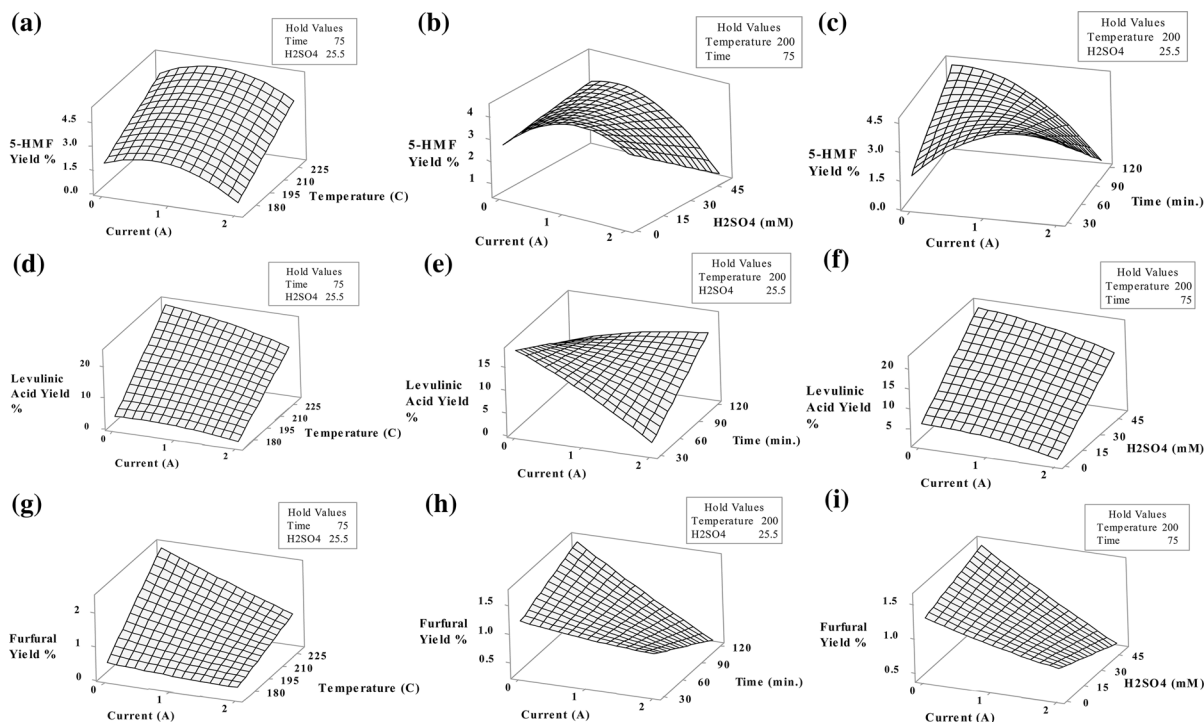


Fig. 9 Surface response plots of 5-HMF, levulinic acid, furfural yields with effecting parameters of applied current, reaction temperature and time, acid concentration (H₂SO₄)

5-HMF, levulinic acid, furfural production

The conversion of cellulose into HMF involves several reactions including hydrolysis to glucose, isomerization of glucose to fructose, dehydration of fructose to HMF (Antal et al. 1990). Further rehydration of HMF resulted in formation of levulinic acid and formic acid. Furfural is also formed by loss of formaldehyde from 5-HMF. There are two possible pathways of fructose dehydration to HMF. One is the cyclic route by the fructofuranosyl intermediate, and the other one is a cyclic route by enol intermediate (Antal et al. 1990). Antal and coworkers investigated the formation of HMF from fructose in H₂SO₄ catalyzed reaction medium of D₂O solvent. They did not observe formation of deuterium-carbon in 5-HMF showing that 5-HMF is formed by acyclic mechanism. Acyclic intermediates can be involved dehydration and fragmentation reactions. In this study, intermediate

products were not considered since GC–MS and HPLC analysis did not detect significant amount of these products.

From the statistical analysis, following equations (Eqs. 7–9) were obtained to calculate the yields of 5-HMF, levulinic acid and furfural ($p < 0.05$).

$$\begin{aligned}
 5-HMF\ Yield\% &= -7.959 + 4.298A + 0.04129B \\
 &+ 0.03203C + 0.01393D \\
 &- 1.139A * A + 0.00646A \\
 &* B - 0.03708A * C - 0.03878A * D
 \end{aligned} \quad (7)$$

$$\begin{aligned}
 Furfural\ Yield\% &= -5.584 + 2.234A + 0.032250B \\
 &+ 0.005500C + 0.00633D \\
 &+ 0.0329A * A - 0.010250A * B \\
 &- 0.006333A * C - 0.006990A * D
 \end{aligned} \quad (8)$$

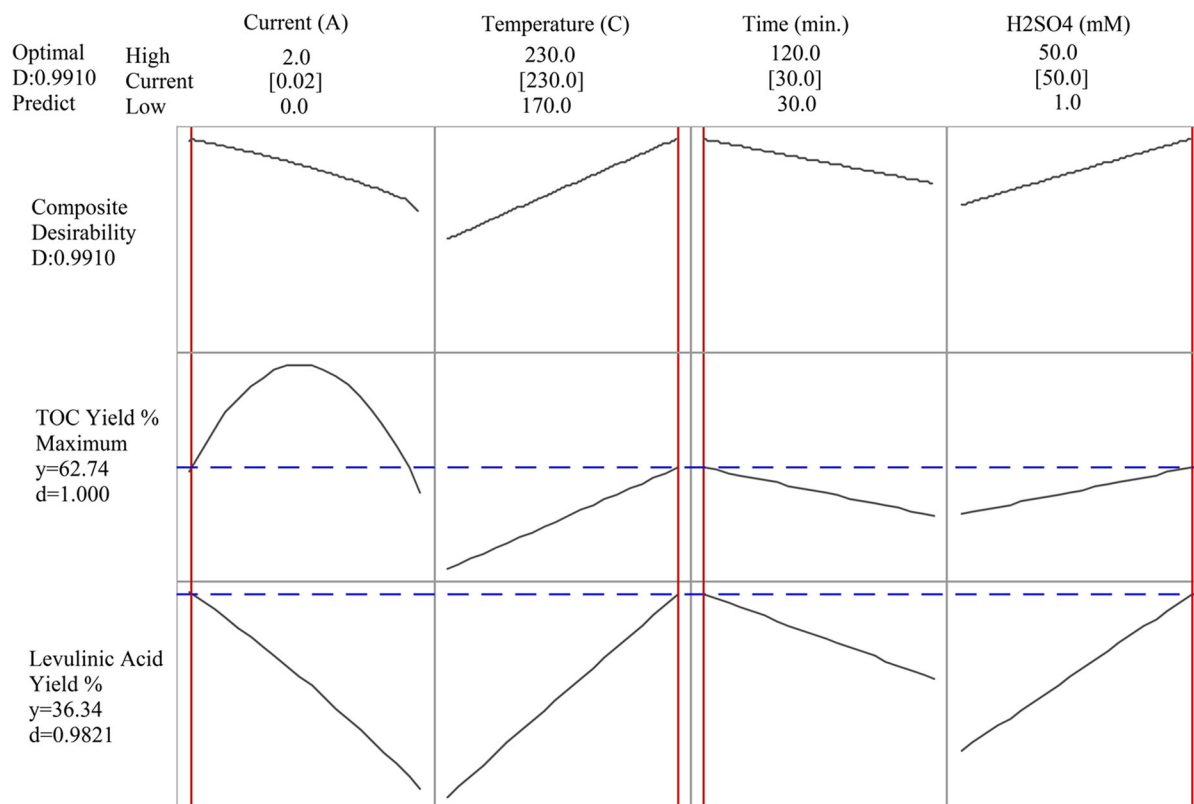


Fig. 10 Optimized parameters for maximum yield of total organic content and levulinic acid

$$\begin{aligned}
 \text{Levulinic Acid Yield\%} = & -55.513 - 0.312A \\
 & + 0.34217B - 0.09800C \\
 & + 0.32510D - 0.918A \\
 & * A - 0.04415A * B \\
 & + 0.11968A * C - 0.02360A * D
 \end{aligned}
 \quad (9)$$

The highest yield of 5-HMF (7 %) was observed when 2 A current was applied at 230 °C for 30 min with the addition of 1 mM H₂SO₄ to the feed solution. Application of current may limit the side reactions of 5-HMF such as polymerization to tarry materials. It was also observed that total organic carbon in the liquid product solution increased by the application of current to the reaction medium. However, the most significant terms in 5-HMF production were found as temperature and H₂SO₄ concentration (Table 4). Response surface plot of 5-HMF yield (Fig. 9b) showed that in current free experiments increase in acid concentration resulted in higher 5-HMF yield. When current was applied after addition of higher acid

concentration (25 mM and 50 mM H₂SO₄), diverse effect in which HMF yield was diminished from 6.84 to 3.73 % was observed. This is due to the further rehydration of 5-HMF to levulinic acid and formic acid. Similar behavior was also observed in current and reaction time interaction (Fig. 9c). At 2 A of applied current experiments, 5-HMF yields at 30 min and 120 min of reaction time were 5.27 and 1.48 %, respectively in contrast with current free experiments, in which reaction time had positive impact on 5-HMF yield; for example 30 min of reaction time yielded 0.95 %, whereas after 120 min the value reached to 3.83 %. This could be due to the formation of acids such as lactic acid, acetic acid and formic acid during the decomposition reaction that changes the ionic constant (K_w) of the reaction medium. Effect of current (2 A) at low reaction time (30 min) supported the idea of electrochemically generated acidic medium near the anode. Because at 2 A of applied current, reaction time had diverse effect on 5-HMF concentration (Fig. 9c) in comparison to levulinic acid

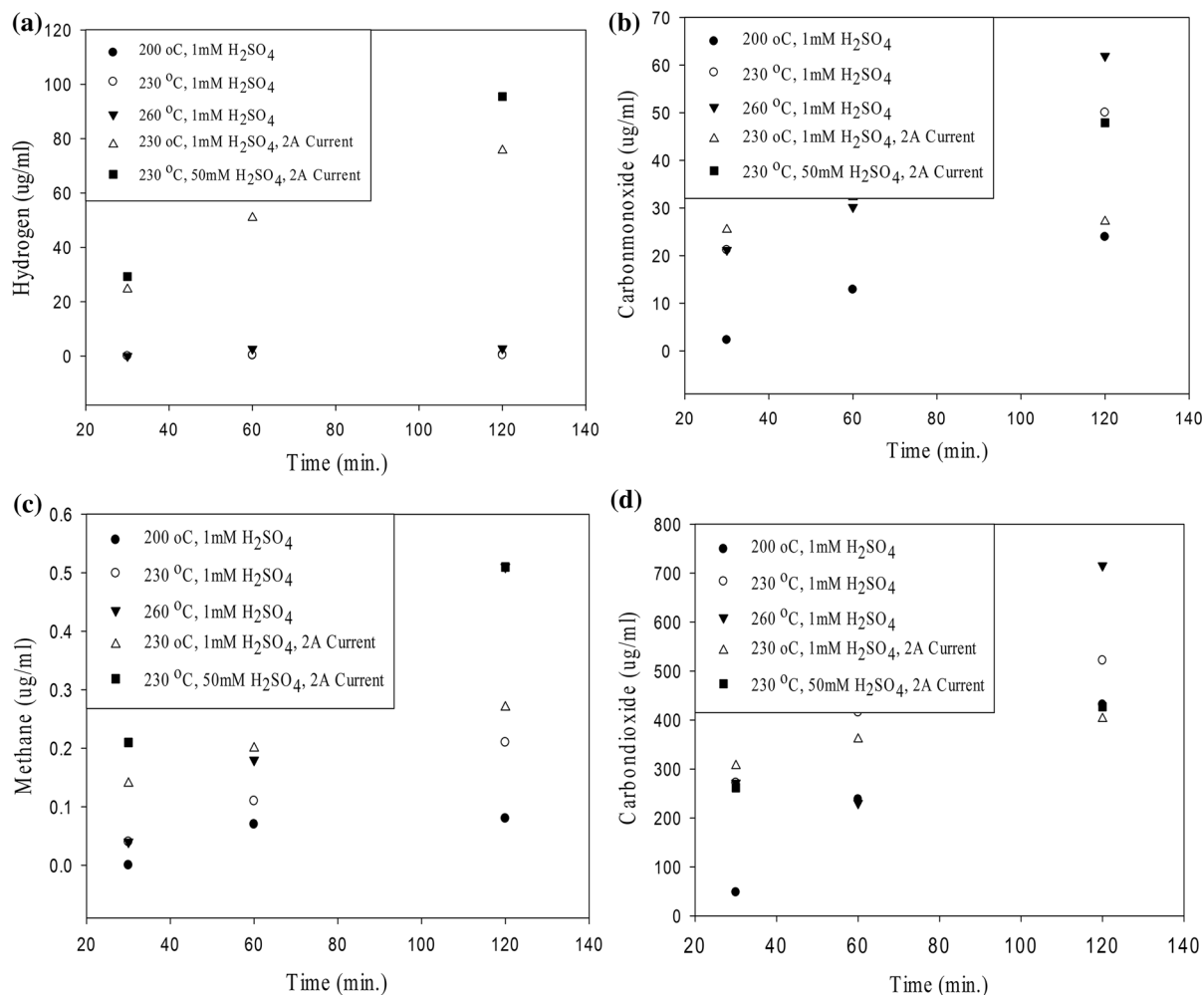


Fig. 11 Formation of gas products **a** hydrogen, **b** carbon monoxide, **c** methane, **d** carbon dioxide during decomposition of cellulose with the effect of applied current at hot-compressed water at different reaction temperatures

concentration (Fig. 9e). Higher H₂SO₄ concentration in applied current experiments resulted in further reactions as rehydration to levulinic acid. Response surface plots of levulinic acid (Fig. 9e) and 5-HMF (Fig. 9c) had the similarity in terms of interaction of applied current and reaction time. Maximum yield of levulinic acid (37 %) was observed without applying current for 30 min at 230 °C with the addition of 50 mM H₂SO₄. Applied current in this conditions resulted in decrease in the levulinic acid yield to 16 %. This decrease in the levulinic acid concentration could be due to restriction of rehydration reaction of 5-HMF under applied current conditions. It was also found that applied current resulted in higher 5-HMF concentration in comparison to current free experiments. Qiu

and coworkers (2014) reported that reduction of levulinic acid in acid solution might result in the formation of 4-hydroxypentanoic acid that yields valeric acid. However, GC–MS analysis did not show the formation of valeric acid, hence, diverse effect of current (1 A) on levulinic acid could be related to the formation of 5-HMF. Reaction time had significant interaction with current and its effect on 5-HMF (Fig. 9c) and levulinic acid (Fig. 9e) concentrations changed with applied current. For instance, when reaction time increased (e.g. 120 min.) at 2 A of current, levulinic acid yield increased from 16 to 28 % and 5-HMF concentration decreased from 4.0 to 0.24 %. However, at current free experiments, after 120 min, levulinic acid yield decreased from 18 to

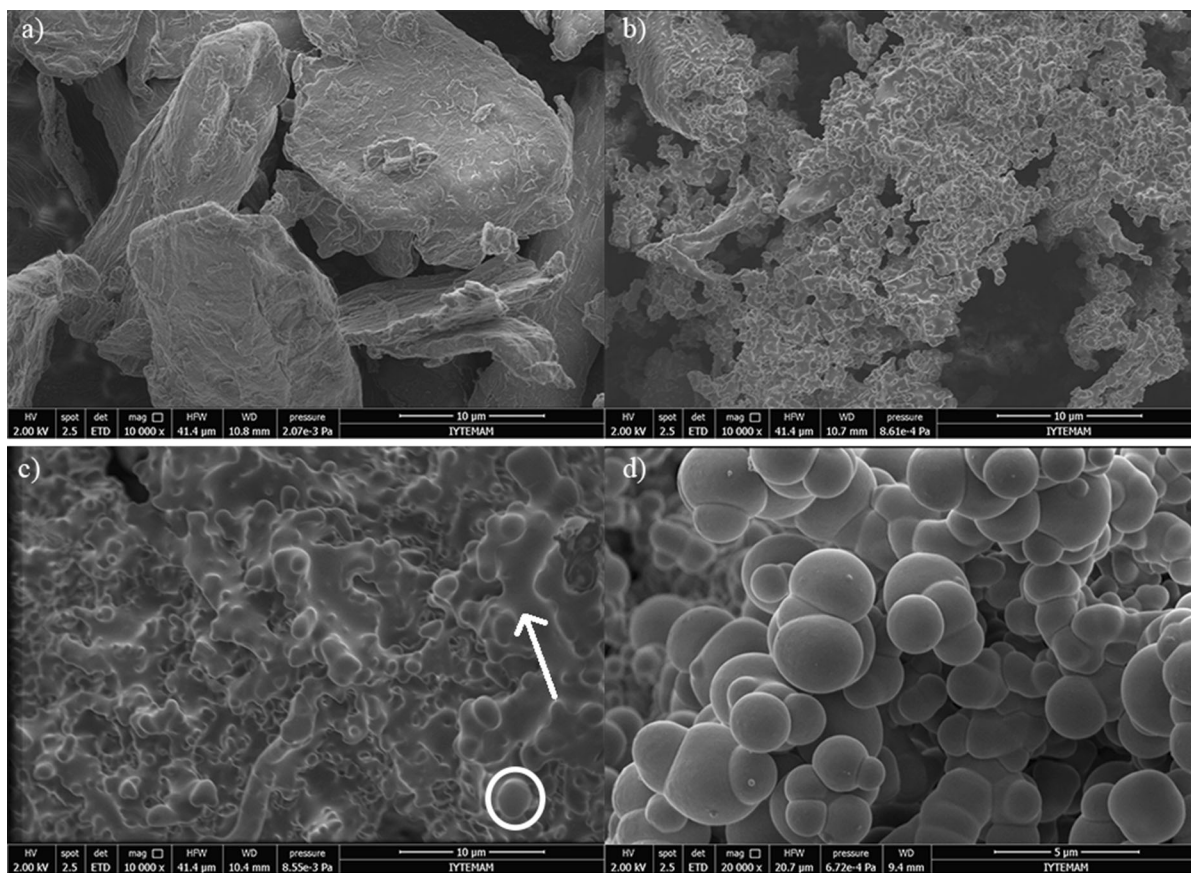


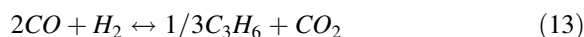
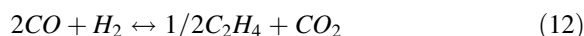
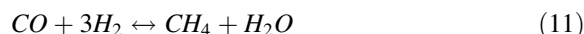
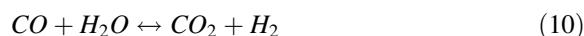
Fig. 12 SEM analysis of **a** microcrystalline cellulose and solid residues of **b** 230 °C, 0 A **c** 260 °C, 0 A and **d** 230 °C after applying 2 A of current (a, b, and c are at magnification of $\times 10,000$ and d is at $\times 20,000$)

9 % and 5-HMF concentration increased from 1.6 to 4.5 %. This replaceable effect of reaction time with current could be due to the change in ionic product concentration, which is affected by electrochemically generated acid layer around anode and formation of degradation products as acids. Figure 10 indicates the optimized parameters to maximize both TOC conversion and levulinic acid yield in the liquid product solution. From this plot, it was predicted that reaction that was carried out with the addition of 50 mM H_2SO_4 by passing 0.02 A current through the electrodes at a reaction temperature of 230 °C would maximize TOC conversion as 62 % and levulinic acid yield as 36 %.

Gas products

Gas samples were collected into plastic bags at sampling times of 30, 60, and 120 min. Collected

samples were analyzed by using gas chromatography thermal conductivity detector. Intermediate products of cellulose degradation such as aldehydes and acids can be further decomposed to carbon monoxide and hydrogen gases under hydrothermal conditions. Possible reactions involved in the formation of gas products as CH_4 , C_2H_4 , C_3H_6 reported as the gas products of cellulose decomposition (Gao et al. 2012) are illustrated as follows;



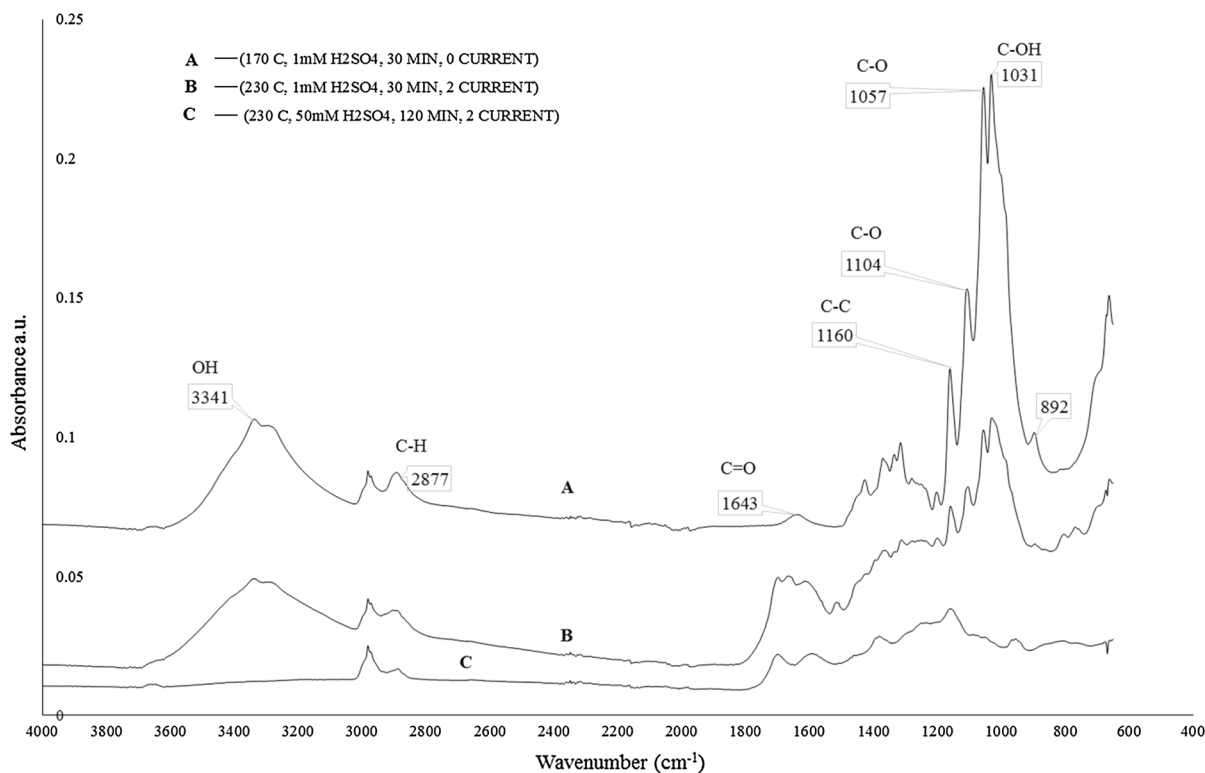


Fig. 13 FTIR spectrum of solid residues at the end of 170 and 230 °C with 2 A current

Figure 11 indicates the formation of gaseous products during decomposition reaction of MCC under hydrothermal conditions. Gas samples were collected during reaction and at the end of the reaction as the medium cooled down to 50 °C. As the reaction temperature increased from 200 °C to 260 °C, hydrogen concentration in the gas products increased from 0.52 to 2.83 $\mu\text{g/ml}$ (Fig. 11a) at the sampling time of 120 min. At higher reaction temperatures, water gas shift reaction favors the hydrogen formation. When 2 A current was applied to the reaction medium, hydrogen concentration reached to 53.8 ($\mu\text{g/ml}$), as shown in Fig. 11a. However, at 1 A current no hydrogen formation was observed. Higher current value can favor the water electrolysis (Eq. 14) that yields in hydrogen and oxygen production.

An increase in hydrogen gas production may lower the rate of protonation of glycosidic bonds of cellulose. 2A of current applied experiments had lower TOC conversion than the experiments carried out under the influence of 1 A current. Therefore, applied current in reaction medium changed the reaction mechanism through the formation of TOC instead of

gas products and char. When 2A current was passed to the reaction medium, the collected gas samples at sampling time of 120 min. and at the end of reaction showed that there was a dramatic decrease in hydrogen gas from 95.48 to 36.71 $\mu\text{g/ml}$ and carbon dioxide gas from 427.14 to 284.83 $\mu\text{g/ml}$ concentrations, respectively. In contrast, carbon monoxide amount increased from 47.89 to 92.24 $\mu\text{g/ml}$. This increase showed that water gas shift reaction (Eq. 10) favored the carbon monoxide side as the reaction temperature cooled from 230 °C to ambient temperature. Methane formation (Fig. 11c) was also observed and an increment in acid concentration from 1 to 50 mM resulted in 0.27 to 0.51 $\mu\text{g/ml}$, respectively.

Analysis of solid residue

Morphological properties of solid residues at the end of cellulose decomposition reactions in hot-compressed water with and without current were analyzed via SEM. MCC is formed by layers (Fig. 12a) of cellulose chains with the particle size of approximately 30 μm . Solid residue with 1 mM H_2SO_4 at 230 °C showed that

cellulose structure was completely destroyed and particles shrunk to lower size (Fig. 12b). At higher temperatures such as 260 °C, when solid residue was monitored by SEM, it was observed that there were solid spheres (circle) formed embedded in tarry materials indicated by an arrow in Fig. 12c. In cellulose degradation, liquid product of 5-HMF can polymerize and forms tarry materials (Chuntanapum et al. 2011). Application of current (2 A, 230 °C) to the reaction medium resulted in the formation of carbon spherical particles (Fig. 12d) with particle size of 1–3 µm. Clear image of microspheres showed that there was no formation of tarry material around. As it was found from statistical analysis results, applied current to the reaction medium would increase the 5-HMF yield in the product solution and would restrict the formation of tarry materials in the solid residue.

The FTIR spectrum of solid residues is shown in Fig. 13. The peaks at 3341 and 2877 cm^{-1} are attributed to stretching vibration of O–H and C–H, respectively. The O–H stretching vibration at 3341 cm^{-1} corresponds to intra-molecular hydrogen bond of cellulose. This peak disappeared at 2 A current and 50 mM H_2SO_4 conditions which indicated that intra-molecular hydrogen bonds were protonated and destroyed. Additionally, the peak at 892 cm^{-1} , which is attributed to β -D-glucopyranosyl, diminished at 230 °C and 2 A of current with reaction time of 30 min. This showed that cellulose hydrolysis reaction resulted in destruction of inter-molecular hydrogen bonds (Chung et al. 2004). Moreover, the broad peak from 1530 to 1750 cm^{-1} shows the existence of aldehydes and ketones absorbed to solid residuals (Gao et al. 2012). This broad spectrum was observed at the residues in which glycosidic bond disappeared. The main characteristic peaks of spectrum 1031 and 1057 cm^{-1} are assigned as C–OH and C–O stretching vibrations. The observation of peaks at 1104 and 1160 cm^{-1} can be attributed to C–O and C–C stretching vibration of cellulose ether, respectively. These peaks were disappeared in residues obtained at 230 °C, 50 mM H_2SO_4 , 2 A of current at 120 min. The peak at 1643 cm^{-1} is assigned as adsorbed water molecules.

Conclusion

Hydrothermal electrolysis experiments of microcrystalline cellulose were carried out under a wide range of

operating parameters by building fractional factorial design. ANOVA test was applied to reveal interaction of applied current with reaction temperature, time and acid concentration. Cellulose conversion reached its maximum value of 82 % at 230 °C, 50 mM H_2SO_4 and 30 min reaction time. In order to reach the maximum conversion value, application of 1 A current decrease the reaction temperature to 200 °C and acid concentration to 1 mM. Moreover, it was found that 1 A applied current to the reaction medium at 200 °C increased the TOC conversion (62 %) by lowering the reaction temperature from 230 to 200 °C and acid concentration from 50 to 1 mM in comparison with current-free experiments. Effect of current may be explained by the formation of acid layer around the anode material resulting in increased hydrolysis rate of glycosidic bond of oligosaccharides. TOC and GC-TCD further supported the idea of electrochemically generated acid layer around anode at lower current values. However, further increase in current (2 A) resulted in decrease of TOC conversion in contrast to an increase in hydrogen production. This can be attributed to increase in hydrolysis reaction rate of water at higher current values.

Levulinic acid concentration reached its maximum yield of 37 % at 230 °C and with 50 mM acid concentration. With the effect of applied current, levulinic acid concentration decreased from 37 to 13 %; however, 5-HMF yield increased to 7 % at 2 A of current applied experiments. As levulinic acid was produced from 5-HMF, applied current may restrict the 5-HMF decomposition.

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