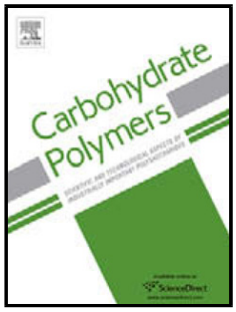


Title: Acid mediated chemical treatment to remove sugar from waste acid stream from nano-crystalline cellulose manufacturing process

Authors: Sampa Maiti, Saurabh Jyoti Sarma, Satinder Kaur Brar, Rama Pulicharla, Richard Berry

PII: S0144-8617(17)30422-8
DOI: http://dx.doi.org/doi:10.1016/j.carbpol.2017.04.033
Reference: CARP 12221

To appear in:

Received date: 23-12-2016
Revised date: 1-4-2017
Accepted date: 14-4-2017

Please cite this article as: Maiti, Sampa., Sarma, Saurabh Jyoti., Brar, Satinder Kaur., Pulicharla, Rama., & Berry, Richard., Acid mediated chemical treatment to remove sugar from waste acid stream from nano-crystalline cellulose manufacturing process.*Carbohydrate Polymers* http://dx.doi.org/10.1016/j.carbpol.2017.04.033

Acid mediated chemical treatment to remove sugar from waste acid stream from nano-crystalline cellulose manufacturing process

Sampa Maiti[a], Saurabh Jyoti Sarma[a], Satinder Kaur Brar[a*], Rama Pulicharla[a], Richard Berry[c]

[a]*Institut National de la Recherche Scientifique (INRS), Centre Eau, Terre & Environnement, 490 de la Couronne, Québec (QC), G1K 9A9, Canada*

[c]*CelluForce, 625-Président-Kennedy avenue, Office 1501, Montreal (Quebec), Canada H3A 1K2*

[1] *Corresponding author: Phone: + 418 654 3116; Fax: + 418 654 2600; E-mail: satinder.brar@ete.inrs.ca*

Abstract

Nano-crystalline cellulose (NCC) is a nano-scale biomaterial derived from highly abundant natural polymer cellulose. It is industrially produced by concentrated acid hydrolysis of cellulosic materials. However, presences of as high as 5-10 % of sugar monomers in spent sulphuric acid during the manufacturing process, makes it unsuitable for such recycling or reuse of sulphuric acid. Currently, the industry has been using membrane and ion exchange technology to remove such sugars, however, such technologies cannot achieve the target of

80-90% removal. In the current investigation, thermal treatment and acid mediated thermal treatment have been evaluated for sugar removal from the spent sulphuric acid. Almost complete removal of sugar has been achieved by this approach. Maximum sugar removal efficiency (99.9%) observed during this study was at 120±1°C for 60 min using 0.8 ratio (sample: acid) or at 100±1°C for 40 min using 1.5 ratio.

Keywords: Acid hydrolysis; nano-crystalline cellulose; sugar removal, Response surface methodology (RSM).

1. Introduction

Cellulose constitutes the most abundant renewable polymer resource available today. Nano-crystalline celluloses (NCCs) have garnered in the materials community a tremendous level of attention that does not appear to be relenting. These bio-polymeric assemblies warrant such attention not only because of their unsurpassed quintessential physical and chemical properties but also because of their inherent renewability and sustainability in addition to their abundance (Habibi, Lucia, & Rojas, 2010). They have been the subject of a wide array of research efforts as reinforcing agents in nano-composites due to their low cost, availability, renewability, light weight, nanoscale dimension, and unique morphology (Peng, Dhar, Liu, & Tam, 2011).

Typical procedures currently employed for the production of NCCs consist of subjecting the pure cellulosic material to strong acid hydrolysis under strictly controlled conditions of temperature agitation, and time (Brinchi, Cotana, Fortunati, & Kenny, 2013; Karim, Chowdhury, Hamid & Ali, 2016; Rhim, Reddy & Luo, 2015). The nature of the acid

and the acid-to-cellulosic fibers ratio are also important parameters that affect the preparation of NCCs (Elazzouzi-Hafraoui et al., 2007; Son & Seo, 2015). A resulting suspension is subsequently diluted with water and washed with successive centrifugations (Bai, Holbery, & Li, 2009). Dialysis against distilled water is then performed to separate the NCC from the acid solution (de Souza Lima & Borsali, 2002). Additional steps such as filtration, differential centrifugation, or ultracentrifugation (using a saccharose gradient) (de Souza Lima & Borsali, 2002) have been also reported (Brinchi et al., 2013; Habibi et al., 2010).

CelluForce® (Montreal, Canada) (http://celluforce.com/en/) is a global leader in nano-crystalline cellulose (NCC^{TM}) production. NCCs manufacturing process of the industry and proposed strategy of removing sugar from the acid solution generated during the process has been presented (Kumar, Negi, Choudhary & Bhardwaj, 2014; Sarma *et al.*, 2016). The diluted sulfuric acid stream generated during NCCs manufacturing process is subjected to membrane based ion exchange technique to remove the remaining sugar monomer. However a significant portion of sugar is still present in the residual acid stream, which could be as high as 5-10%. Owing to strong hygroscopic nature of sulfuric acid, it makes it difficult for the manufacturer to concentrate and reuse the same acid stream in presence of such sugars during a subsequent batch. Currently, waste acid stream from manufacture industry is utilized mainly to produce biogas or sent to wastewater treatment facility at additional expense. Thus, in order to make the process more sustainable and environmentally friendly, the industry has been looking for a simple method to remove at least 80-90% of the sugar without neutralizing the acid solution, so that it can be concentrated for reuse. Therefore, development of a new cost-effective and efficient process for the removal of at least 80-90 % of such sugar was the major objective of this study.

During acid catalyzed thermal-hydrolysis, biopolymeric assemblies (e.g. cellulose and hemicellulose) are degraded into monomers (e.g. hexoses and pentoses) and further byproducts as shown in Figure-1 (Choudhary et al., 2013; Shen, Zhang, Xue, Guan, Liu & Xiao, 2015). However, depending on the process conditions, the conversion of these biopolymers could be directed in different directions, such as sugars monomers (e. g: glucose) (Rugg & Brenner, 1982); furfural derivative (furfural and 5-hydroxymethyl furfural) (Choudhary et al., 2013); levulinic acid (Girisuta, Janssen, & Heeres, 2007), nanostructured ceramics and nano-composites (Pang, Chin, & Yih, 2011) and others via rehydration, dehydration and other mechanism (Choudhary et al., 2013). In this context, sulfuric acid mediated heating could be a promising novel technology for sugar removal from diluted sulfuric acid stream generated during nano-crystalline cellulose (NCC^{TM}) manufacturing process. Hence, the objectives of the study were: (1) physicochemical characterization of waste acid stream from NCC manufacturing process; (2) heating at different temperatures to evaluate the effect of heating on sugar removal; (3) evaluation of acid mediated heating using different ratios of sample and acid as a novel method for sugar removal and; (4) optimization of process parameters, such as time, temperature and acid sample ratio to enhance sugar removal process. Central composite design was used for optimizing these parameters. This article will provide an alternative solution to remove residual sugar from the waste acid stream from NCC^{TM} manufacturing process and the proposed method has the potential to be an environment-friendly solution.

2. Materials and methods

2.1 Chemicals and Waste acid stream sample

Chemicals, such as sulfuric acid, sodium hydroxide, acetone, methanol, acetonitrile, NH_4OH etc. were purchased from Fisher Scientific (Ontario, Canada). Glucose, xylose, fructose, treholose, furfural, hydroxymethyl furfural, levulinic acid, acetic acid, among others have been purchased from Sigma Aldrich (USA). All the standards used for analytical methods are of analytical grade. Waste acid stream from NCC manufacturing process was received from CelluForce® (Windsor, Quebec).

2.2 Heating experiment on sugar removal from waste acid stream of manufacturing process

Heat mediated in-situ acid catalyzed the hydrolysis of cellulose was performed to reduce sugars and further conversion of other byproducts to reuse the acid. For this investigation, heating at different temperatures, such as 40 ºC, 60 ºC, 80 ºC and 100 ºC have been carried out using about 10 mL of sample from the sulfuric acid stream received from CelluForce® (Windsor, Quebec). Each sample was heated to the aforementioned temperatures in a closed COD vial and boiled for 120 minutes. After heating, each sample was cooled to room temperature and total reducing sugars, total carbohydrates and byproducts were analyzed using different analytical methods (Maiti et al., 2016; Miller, 1959) (http://web.itu.edu.tr/~dulekgurgen/Carbs.pdf).

2.3 Acid mediated heating experiment on sugar removal from waste acid stream of manufacturing process

During the transformation of the biopolymer, such as cellulose to sugar monomer and other byproducts, several operational variables interact and influence the process (Zhang, Xin, Liu, & Ge, 2015). Slight change in process condition could lead the entire process in a definite direction (Choudhary et al., 2013). Thus, in order to enhance the sugar removal and by-products removal, different ratios of (sample: sulfuric acid) have been used at different temperatures as mentioned earlier in a closed COD vial and boiled for 120 minutes. After heating, each sample was cooled to room temperature and total reducing sugars, total carbohydrates and byproducts were analyzed using different analytical methods. Sugar was finally converted to black nanoparticles. These particles were removed from the solution by the method described in section 3.7.

2.4 Experimental design and sugars and byproducts removal optimization through response surface methodology (RSM)

During acid hydrolysis of cellulose, several operational variables interact and influence the response. Determination of the optimum point can be achieved with a limited amount of experiments through statistical analysis. Response surface methodology (RSM), a compilation of mathematical and statistical techniques, can be used to interpret and evaluate the combined effects of all the factors in the hydrolysis process (Zhang et al., 2015). Central composite design (Zhang et al., 2015) was applied to investigate sugars and byproducts removal (dependent variable) as a function of three independent variables: reaction time (A), temperature (B) and sample & concentrate sulphuric acid ratio (C). Design-Expert-7 software (Stat-Ease Inc. Minneapolis, MN) has been used to construct the experimental design and the design has been extended up to $+\alpha$ and $-\alpha$ level (Maiti et al., 2015). The experimental design resulted in a set of 20 experiments, comprising three different code levels (low (-1), middle

(0) and high (+1)). The ranges of the variables investigated and responses were reported in terms of sugars (glucose, xylose, and trehalose) and by-products (levulinic acid, 5-HMF) removal as shown in Table-1 and Table-2.

A quadratic polynomial equation (Eq. 1) was proposed to interlink the effects of the three independent variables on sugar removal as follows:

$$(\text{Sugar/Byproducts})_{\text{removal}} = X_0 + \sum_{i=1}^{n} X_i Y_i + \sum_{i=1}^{n} X_{ii} Y_i^2 + \sum_{i=1}^{n-1} \sum_{j=i+1}^{n} X_{ij} Y_i Y_j$$

(Eq. 1)

where, $(\text{Sugar/Byproducts})_{\text{removal}}$ is the dependent variable; Y_i and Y_j are the independent variables (n = 3 (A, B and C); X_0 is the intercept constant and X_i, X_{ii} and X_{ij} correspond to the regression coefficients. The same software (Design-Expert®-7) employed for test-matrix design was used to check the experimental responses obtained. An analysis of variance (ANOVA report) was performed to fit the quadratic polynomial equations. Final values of code factors (A, B, C, A^2, B^2, C^2 etc.) were considered to be statistically significant at $p< 0.05$. The quality of the model fit was evaluated by the coefficient of determination (R^2) and the adjusted coefficient of determination (R^2_{Adj}).

2.5 Total reducing sugar analysis by standard DNS method

Total reducing sugar concentration was analyzed by di-nitro salicylic acid (DNS) method (Miller, 1959). Briefly, 200 µL solutions of the sample were taken each time. It was mixed with 800 µL water and 2 mL of 3, 5 di-nitro salicylic acid reagents (alkaline) and placed in water bath for 10 minutes at 100 ± 1ºC and cooled to room temperature. Later, about 7 mL water was added and the solution was vortexed for 30 sec. The optical density

was measured at 540 nm using 200 µL of resulting solution in 96 well polystyrene assay plate and absorbance readings were then compared to a standard curve prepared using standard glucose solution.

2.6 Determination of sugar concentration in the acid stream

Sugar concentration in the acid stream was determined by anthrone method and expressed as total carbohydrate. Briefly, the sample was diluted 50 times with distilled water and 1 mL of diluted sample was taken. Later, about 2 mL of chilled H_2SO_4 (75% v/v) was mixed with the sample. This solution was then mixed with 4 mL of chilled anthrone reagent and the resulting mixture was taken in a closed COD vial and boiled for 15 minutes at 100 ±1 ºC. After cooling to room temperature, absorbance was measured at 578 nm (http://web.itu.edu.tr/~dulekgurgen/Carbs.pdf). Anthrone reagent used in this analysis was freshly prepared by dissolving 0.5 g of anthrone in 10 mL of ethanol and by making up the volume to 250 mL by using H_2SO_4 (75%).

2.7 Reducing sugars and different by-products analysis by LC/MS-MS method

Reducing sugars (e.g: glucose etc.) and complex mixture of other by-products (such as furfural, 5-hydroxymethyl furfural (5-HMF), levulinic acid, among others) were generated during acid hydrolysis of cellulose. To analyze different reducing sugars, liquid samples were collected and analyzed using Liquid Chromatography - Mass Spectrometry equipped with 5µm, 150 mm ID, 4.6 mm df column where D_6 glucose was used as internal standard. Likewise, different inhibitors produced during hydrolysis were analyzed by Liquid Chromatography - Tandem Mass Spectrometry (ZORBAX Carbohydrate, Agilent Technologies, USA) equipped with biobasic-18 column (5µm, 250 mm ID, 4.6 mm df) of Agilent Technologies, USA was used. Before injecting the sample in LC/MS-MS for product analysis, the sample was centrifuged for 5 minutes at 7650 *x g* and the supernatant was

filtered by 0.45 µm syringe filter. Phenylethanol-D_5 is used as internal standard for inhibitory analysis. Before injecting the sample in LC/MS-MS for product analysis, the sample was centrifuged for 5 minutes and the supernatant was filtered by 0.45 µm syringe filter. Methanol: water (8:2) and acetonitrile: water (8.5:1.5) was used to dilute the sample before analyzing the inhibitors and carbohydrate. All data presented are an average value from duplicate runs for triplicate samples. The standard deviation for each data has been calculated with respect to the average (mean) value from duplicate runs for triplicate samples.

Sugars, as well as by-products removal,, were calculated using (Eq. 2):

$$\text{Removal of (sugar/ byproducts) (\%)} = 100 \times \frac{\text{Sugar/ byproducts acid removed after reaction (g)}}{\text{Initial sugars/ byproducts (g)}} \quad \text{(Eq. 2)}$$

2.8 Particle size analysis

Black particles obtained during the current investigation were characterized for particle size and zeta-potential distribution. This analysis was carried out using a zetasizer nano ZS (Malvern instruments Ltd., UK). Distilled water was used to disperse the activated carbon particles used in this analysis. Finally, scanning electron microscope (SEM) analysis was carried out for further investigation.

2.9 Statistical analysis

All the data were an average of successive three reading of measurements using either standard or developed methods. Standard deviations were calculated based on these means and average values.

3. Results and discussion

3.1 Characterization of waste acid stream sample

Prior to sugar removal experiment, physicochemical characterization of the waste acid stream from manufacturing process has been analysed to assess carbon pool. The pH of the waste acid stream sample was about (0.6±0.1) whereas its total carbohydrate concentration was about 6510 mg/L. Glucose (3664.5 mg/L) and xylose (857.1 mg/L) were the two major sugar monomers present in the solution. A small amount of disaccharides, such as trehalose (102.4 mg/L) and oligosaccharides were the other likely constituents of the sample together with by-products, such as furfural derivatives, furfural (56.8 mg/L), 5-hydroxy methyl furfural (110.2 mg/L) and levulinic acid (76 mg/L).

3.2 Heating experiment on sugar removal from waste acid stream from manufacturing process

During acid catalyzed thermal-hydrolysis, cellulose degraded into hexose (e.g. glucose). Hexoses are primarily dehydrated to 5-HMF, which is accelerated by Brønsted acid catalysts, and thereupon 5-HMF is rehydrated into levulinic acid as shown in Figure-1.However, depending on the process conditions, these sugar monomers could also end up with other by-products rather than levulinic acid (Choudhary et al., 2013; Galletti, Antonetti, De Luise, Licursi, & Nassi, 2012; Hu, Song, Wu, Gholizadeh, & Li, 2013). Typically, the concentration of sulphuric acid during hydrolysis reactions to obtain NCCs does not vary much from a typical value of ca. 65% (wt); however, the temperature can range from 25 °C up to 70 °C and the corresponding hydrolysis time can be varied from 30 min to overnight depending on the temperature (Habibi et al., 2010). In order to study the in-situ acid catalyzed conversion of free sugar monomers to corresponding by-products, the samples were heated at different temperatures as mentioned in section 2.2. The purpose of this study was to convert sugar monomer to other by-products to remove sugar from the waste sample as shown in Figure-1. However, no significant colour change and by-products conversion was observed at the end of experiments as shown in Figure-2 (a). From Figure -2 (a), it has been evident that

with increase in heating temperature (40-100 °C), percentage of removal is slowly increased and compared with control, percentage of sugar removal from heat mediated auto-hydrolysis leads to only around (15-20%) from the waste effluent at 100°C for 1 hr. Moreover, to enhance this removal efficiency, different time durations have been considered for further investigation. Percentage of sugar removal with increased time duration was increased but ranged from (20-30%). Thus, heating up to 100 °C for different time durations in closed COD tube was not so effective to provide enough activation energy for higher conversion of reducing sugar to other by-products.

3.3 Acid mediated heating experiment on sugar removal from waste acid stream from manufacturing process

Effect of diluted different mineral acid on hydrolysis has been investigated previously by (Sarchami el at., 2014). It has been reported that as compared to HCl, H_3PO_4, sulfuric acid (H_2SO_4) had a positive impact on diluted mineral acid catalyzed the hydrolysis of cellulose (Hamid & Ali, 2016; Sarchami & Rehmann, 2015). Thus, for the current investigation, H_2SO_4 has considered for dilute mineral acid catalyzed the hydrolysis of the waste sample. Previously, it has been observed that the sugar removal efficiency slightly increases with an increase in temperature. Thus, acid mediated thermal treatment was carried out initially at 100°C for 2 h using different ratios of sample: sulphuric acid concentrated (H_2SO_4: 18M). As mentioned in the materials and methods section, the purpose of this study was to hydrolyze any disaccharides or oligosaccharides which were present in the solution to corresponding monomers as well as the further conversion of monomer to other by-products to remove the sugars from the solution. Different ratios of the sample and sulfuric acid have been investigated to enhance the removal efficiency of sugar from waste effluent. Results of

this investigation have been presented in Figure-2 (b). From Figure-2 (b), it has been shown that increase in the sample: acid ratio had a positive effect on the removal of sugar from waste effluent. About (1:1) ratio of sample and sulphuric acid and higher (sulphuric acid: sample) proved to be promising compared to other ratios in current process condition. Around 99% of sugar (glucose, xylose trehalose) removal was observed when (1:1) ratio of sample and 18 (M) sulphuric acid was mixed and heated together at 100°C for 2 h. Thus, present process conditions ((1:1) ratio of sample and sulphuric acid, 100°C for 2 h) was promising for removal of sugar from the waste sample.

3.4 Optimization of process parameters to enhance sugar removal efficiency from waste acid stream from manufacturing process

The responses (sugars removal from the waste acid stream from the manufacturing process) obtained for different runs of experiments carried out using four independent variables (reaction time (A), temperature (B) and the ratio of sample and sulphuric acid (C) have been presented in Table-2. Obtained responses of the experiments were analyzed by using the same software (Design-expert) used for designing of the experiments. The response surface quadratic model was suggested for the response observed in terms of glucose, xylose and trehalose removal. The ANOVA reports for the response surface quadratic model are presented in Table-3. The model was highly significant ($p < 0.0004$) for the observed responses of sugar removal from the waste acid stream from the manufacturing process. In fact, smaller the magnitude of p ($p< 0.05$), the more significant the corresponding factor. Based on this criterion, temperature (B) and sample sulphuric acid ratio (C) showed a significant effect ($p< 0.05$) in LA production. On the other side, time (A) presented comparatively less significant effect on LA production. The quadratic terms, such as (substrate: sulphuric acid) showed a highly significant negative effect on sugar removal ($p< 0.0001$) indicating the fact that it could turn into a limiting factor and its variation could

lower sugar removal. Corresponding equations to predict sugar removal from the waste acid stream from the manufacturing process in terms of real factors are as follows (Eq. 3- 4):

$$Glucose_{removal} = -362.86 + 0.79 \times Time + 3.21 \times Temp + 282.98 \times H2SO4 - 4.49E\text{-}003 \times Time \times Temp - 0.128 \times Time \times H2SO4 - 1.34 \times Temp \times H2SO4 - 5.29E\text{-}004 \times Time^2 - 34.81 \times H2SO4^2 - 1.196E\text{-}003 \times Temp^2 \quad \text{(Eq.-3)}$$

$$Xylose_{removal} = -225.83 + 0.79 \times Time + 3.21 \times Temp + 282.98 \times H2SO4 - 4.49E\text{-}003 \times Time \times Temp - 0.128 \times Time \times H2SO4 - 1.34 \times Temp \times H2SO4 - 5.29E\text{-}004 \times Time^2 - 34.81 \times H2SO4^2 - 1.196E\text{-}003 \times Temp^2 \quad \text{(Eq.-4)}$$

The goodness of the model adjusted for the range of variables posed was checked by the determination coefficient (R^2). In both models, R^2 values higher than 0.93 indicated that 93% variations in sugar removal can be well explained by the model. The adjusted R^2 value higher than 93% indicated that experimental and predicted values fit well. Sugar removal ranged from about 6.18% to a maximum 99.99% for glucose, while it oscillated from 48.98% g to 99.99 and 81.20% to 99.99% for xylose and trehalose removal.

Figure 3 (a) and 3 (b) show the response surface plots and the corresponding contour plot of the quadratic model developed for sugar removal, such as glucose and xylose, respectively from the waste acid stream from the manufacturing process. Each figure consists of three graphs, where one different variable is kept at a constant level and the other two varied with the experimental values chosen. Therefore, the relationship between sugar

removal and experimental levels of each variable and interactions between the remaining two independent terms are visually observed. The shapes of the plots are related to the intensity of interactions between the independent terms (Zhang et al., 2015). In this case, the three-dimensional surface diagram given for the both substrates indicated strong interactions between all three of them. Additionally, a local optimum point was defined for each case in the experimentally investigated range, which indicated that sugars removal were sensitive to modifications in the independent variables' values.

Regarding the optimized parameters, currently optimized acid mediated heating allowed 50% reduction in processing time from 2 h to 1 h. Apart from process time reduction, the heating method has offered additional improvements (e.g. sample: acid ratio reduction) in the literature (Szabolcs, Molnár, Dibó, & Mika, 2013). With respect to sample: acid ratio, sugar removal achieved a maximum at 0.8 for all sugars, such as glucose, xylose, and trehalose as shown in Figure-3 (a) and (b). Even for the current investigation, the addition of higher ratio compared to 0.8 also resulted in the same (99.9% removal of each sugar). These results were similar to the literature when enhanced acid strength mediated hydrolysis of biopolymers (such as cellulose, hemicellulose) resulted in higher levulinic acid production with no sugar remaining (Galletti et al., 2012). A temperature setting of 120±1 °C was chosen based on current investigation and to avoid the sharp increase of brown/black humins generation (dehydration undesired by-products) when reaction temperature was above 170 °C which was hard to separate (Szabolcs et al., 2013).

Thus, it is evident that acid mediated heating at 120°C, 0.8 ratio, for 60 min removed more than 99% of sugars from the waste sample. However, with an increase in the sample: acid ratio, similar results were also obtained at a comparatively lower temperature of around 100°C and lower time of 40 min as shown in Figure 4 and 5. Furthermore, either heating at

120°C for 60 min using 0.8 ratio or at 100°C for 40 min using 1.5 ratio were observed to be promising options sugar removal from the waste sample.

3.5 Investigation of different by-products during acid mediated heating experiment on sugar removal from waste acid stream from manufacturing process

During acid mediated hydrolysis of biopolymer (e.g. cellulose) monomer, such as glucose was produced and depending on the process condition was further converted to different by-products (e.g. furfural, 5-hydroxy methyl furfural, levulinic acid, among others) as shown in Figure-1. Results of this investigation have been presented in Figure 6. As seen earlier, increase in the sample: acid ratio had a positive effect on by-products formation. With the increase in the ratio (sample: acid) from (6:1) to (1:1) by-products, levulinic acid production significantly increased from 0.19 g/L to 2.1 g/L. Previously, it has been observed that when (1:1) ratio was used, around 99% sugar was removed which implied that this sugar has been converted to mainly levulinic acid and other by-products as shown in Figure-4. Although, this method can convert sugar to its derivatives, still it cannot be considered as actual sugar removal as long as the by-products are there. However, for the same experimental conditions, when the ratio was changed to (1:1.5) no sugar as well as by-products, such as levulinic acid, furfural, 5-hydroxymethyl furfural was reported (Figure-4). Thus, (1:1.5) sample: acid ratio led to around 99.99% of sugar and by-products removal from the waste acid solution. All these compounds were converted to black carbon particles which were removed by centrifugation. Furthermore, different parameters, such as time, temperature and sample: acid ratio which have their influence in by-products formation, have been optimized using central composite model (Zhang et al., 2015).

3.6 Optimization of process parameters to enhance by-products removal from waste acid stream from manufacturing process

The similar central composite design was used to study the interaction effects of the three factors and find the suitable values of these variables to remove by-products and the measured responses are mentioned in session 3.1. Results of this study have been presented in Figure-S1 (supplementary file) Increase in substrate acid ratio and temperature have a positive effect on by-products, such as levulinic acid and 5-HMF removal. Thus, at 100°C for 40 min using 1.5 sample: acid ratio was observed to be promising in by-products removal from the waste sample.

3.7 Investigation of black particles formed at the end of acid mediated heating experiment on sugar removal from waste acid stream from manufacturing process and reuse of acid

Black particles were produced during the treatment to remove sugar present in the waste acid stream. To analyze the black particles produced at the end of acid mediated heating, particle size and zeta-potential distribution were measured. Particles are found to be around 18.6 nm in diameter which indicated that present black particles were of nano-diameter. For further characterization of the particles, scanning electron microscope (SEM) imaging was carried out as shown in Figure-5. Figure-5 (A-1 and A-2) represented the SEM image of starting material and the particle size was about 1.5 µm. Figure-5 (B-1 and B-2) represented the SEM of the sample after acid treatment and containing the black particles. From the images, it is evident that the black particles are of needle shape and with nano-diameter. However, in order to reuse the acid, removal of black particles was required. To remove the black nanoparticles generated during treatment, sodium hydroxide treatment was carried out. Around 0.1 (M) of NaOH was used in different ratios with sample followed by

centrifugation. From the results, it was evident that (sample: 0.1 (M) NaOH = 1:1) ratio was promising in this context followed by centrifugation. Thus, acid mediated heating treatment followed by NaOH treatment could assist in the reuse of acid instead of biogas production or being sent to wastewater treatment facility at additional expense.

4. Conclusions

Acid mediated thermal treatment of waste acid stream from NCC manufacturing process was confirmed to be promising in sugar removal. The approach will be helpful for efficient recycling of the acid used in the process. An investigation using response surface methodology has shown that temperature, time and substrate acid ratio are the dominating factors for successful removal of sugar as well as different byproducts. Heating at 120°C for 60 min using 0.8 sample: acid ratio or at 100°C for 40 min using 1.5 sample: acid ratio showed excellent results in sugar or and by-products removal from the waste sample.

5. Acknowledgement

NSERC CRD and Prima-Quebec have been acknowledged for financial support. Contributions of the technical staff of INRS involved in sample analysis have been acknowledged. CelluForce is acknowledged for providing sugar samples and technical support.

Reference

Bai, W., Holbery, J., & Li, K. (2009). A technique for production of nanocrystalline cellulose with a narrow size distribution. *Cellulose, 16*(3), 455-465.

Brinchi, L., Cotana, F., Fortunati, E., & Kenny, J. (2013). Production of nanocrystalline cellulose from lignocellulosic biomass: technology and applications. *Carbohydrate Polymers, 94*(1), 154-169.

Choudhary, V., Mushrif, S. H., Ho, C., Anderko, A., Nikolakis, V., Marinkovic, N. S., Frenkel, A. I., Sandler, S. I., & Vlachos, D. G. (2013). Insights into the interplay of Lewis and Brønsted acid catalysts in glucose and fructose conversion to 5-(hydroxymethyl) furfural and levulinic acid in aqueous media. *Journal of the American Chemical Society, 135*(10), 3997-4006.

de Souza Lima, M. M., & Borsali, R. (2002). Static and dynamic light scattering from polyelectrolyte microcrystal cellulose. *Langmuir, 18*(4), 992-996.

Elazzouzi-Hafraoui, S., Nishiyama, Y., Putaux, J.-L., Heux, L., Dubreuil, F., & Rochas, C. (2007). The shape and size distribution of crystalline nanoparticles prepared by acid hydrolysis of native cellulose. *Biomacromolecules, 9*(1), 57-65.

Galletti, A. M. R., Antonetti, C., De Luise, V., Licursi, D., & Nassi, N. (2012). Levulinic acid production from waste biomass. *BioResources, 7*(2), 1824-1835.

Girisuta, B., Janssen, L., & Heeres, H. (2007). Kinetic study on the acid-catalyzed hydrolysis of cellulose to levulinic acid. *Industrial & engineering chemistry research, 46*(6), 1696-1708.

Habibi, Y., Lucia, L. A., & Rojas, O. J. (2010). Cellulose nanocrystals: chemistry, self-assembly, and applications. *Chemical reviews, 110*(6), 3479.

Hamid, S. B. A., Chowdhury, Z. Z., Karim, M. Z., & Ali, M. E. (2016). Catalytic Isolation and Physicochemical Properties of Nanocrystalline Cellulose (NCC) using HCl-FeCl3 System Combined with Ultrasonication. *BioResources, 11*(2), 3840-3855.

Hu, X., Song, Y., Wu, L., Gholizadeh, M., & Li, C.-Z. (2013). One-pot synthesis of levulinic acid/ester from C5 carbohydrates in a methanol medium. *ACS Sustainable Chemistry & Engineering, 1*(12), 1593-1599.

Karim, M. Z., Chowdhury, Z. Z., Hamid, S. A., & Ali, M. E. (2016). Optimizing the pretreatment condition for α-cellulose using lewis acid catalyst. *Sci. Adv. Mater., 8*, 534-544.

Kumar, A., Negi, Y. S., Choudhary, V., & Bhardwaj, N. K. (2014). Characterization of cellulose nanocrystals produced by acid-hydrolysis from sugarcane bagasse as agro-waste. *Journal of Materials Physics and Chemistry, 2*(1), 1-8.

Maiti, S., Sarma, S. J., Brar, S. K., Le Bihan, Y., Drogui, P., Buelna, G., & Verma, M. (2016). Agro-industrial wastes as feedstock for sustainable bio-production of butanol by Clostridium beijerinckii. *Food and Bioproducts Processing, 98*, 217-226.

Maiti, S., Sarma, S. J., Brar, S. K., Le Bihan, Y., Drogui, P., Buelna, G., Verma, M., & Soccol, C. R. (2015). Novel spectrophotometric method for detection and estimation of butanol in acetone–butanol–ethanol fermenter. *Talanta, 141*, 116-121.

Miller, G. L. (1959). Use oi Dinitrosalicylic Acid Reagent tor Determination oi Reducing Sugar. *Analytical chemistry, 31*(3), 426-428.

Pang, S. C., Chin, S. F., & Yih, V. (2011). Conversion of cellulosic waste materials into nanostructured ceramics and nanocomposites. *Adv. Mater. Lett, 2*(2), 118-124.

Peng, B. L., Dhar, N., Liu, H., & Tam, K. (2011). Chemistry and applications of nanocrystalline cellulose and its derivatives: a nanotechnology perspective. *The Canadian Journal of Chemical Engineering, 89*(5), 1191-1206.

Rhim, J.-W., Reddy, J. P., & Luo, X. (2015). Isolation of cellulose nanocrystals from onion skin and their utilization for the preparation of agar-based bio-nanocomposites films. *Cellulose, 22*(1), 407-420.
Rugg, B. A., & Brenner, W. (1982). Process for the chemical conversion of cellulose waste to glucose. Google Patents.
Sarchami, T., & Rehmann, L. (2015). Optimizing acid hydrolysis of Jerusalem artichoke-derived inulin for fermentative butanol production. *BioEnergy Research, 8*(3), 1148-1157.
Shen, D., Zhang, L., Xue, J., Guan, S., Liu, Q., & Xiao, R. (2015). Thermal degradation of xylan-based hemicellulose under oxidative atmosphere. *Carbohydrate Polymers, 127*, 363-371.
Son, H. N., & Seo, Y. B. (2015). Physical and bio-composite properties of nanocrystalline cellulose from wood, cotton linters, cattail, and red algae. *Cellulose, 22*(3), 1789-1798.
Szabolcs, Á., Molnár, M., Dibó, G., & Mika, L. T. (2013). Microwave-assisted conversion of carbohydrates to levulinic acid: an essential step in biomass conversion. *Green Chemistry, 15*(2), 439-445.
Zhang, Y., Gao, X., Liu, J., & Ge, Y. (2015). Pilot production of Clonostachys rosea conidia in a solid- state fermentor optimized using response surface methodology. *Engineering in Life Sciences, 15*(8), 772-778.

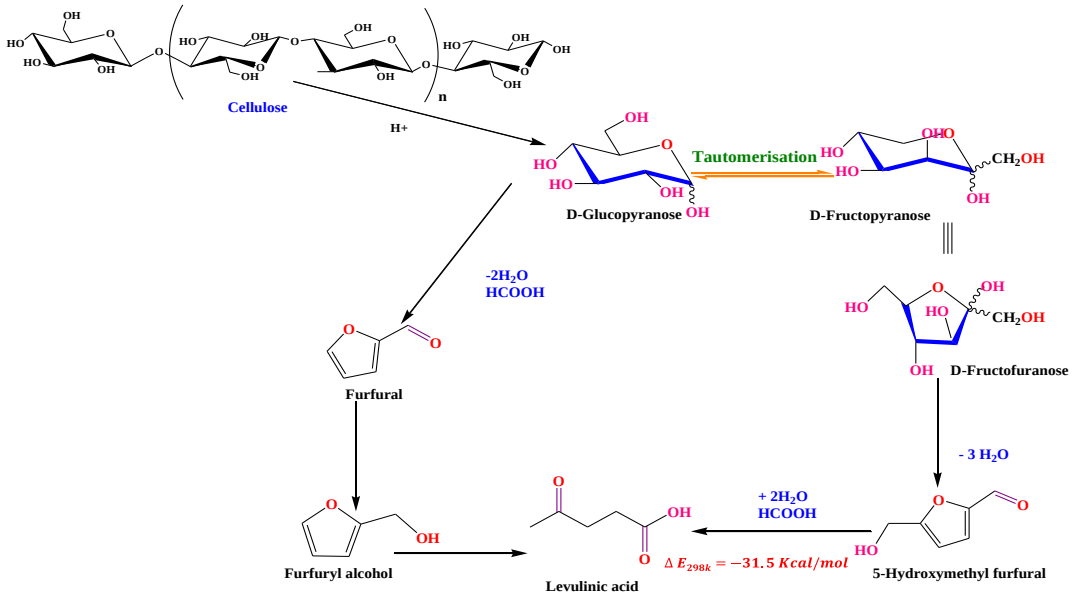
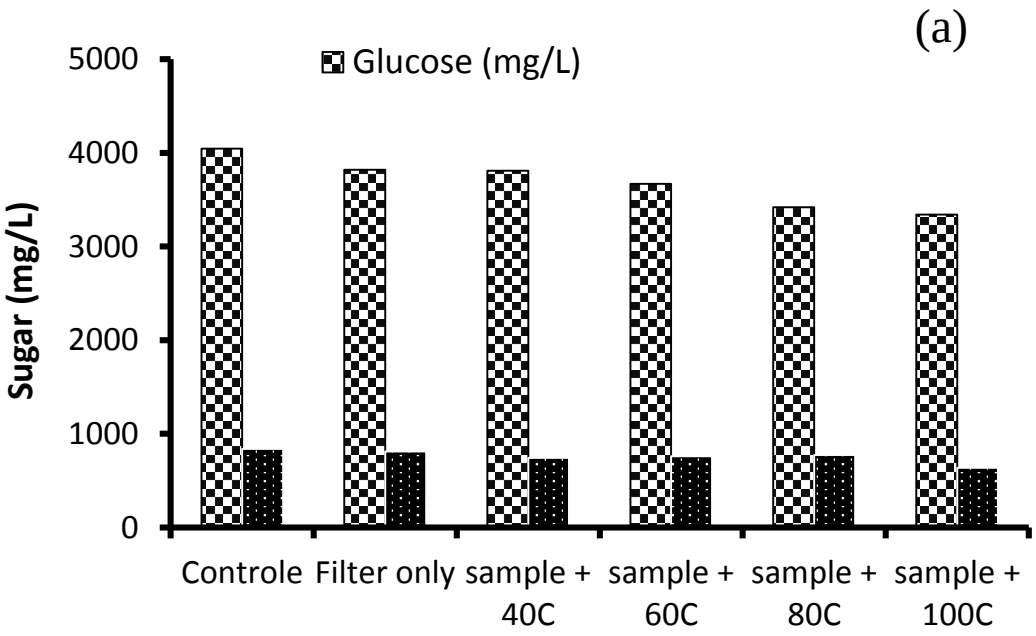


Figure-1: Schematic presentation of acid catalyzed conversion of nanocrystalline cellulose (NCCs) to monomer (e.g. glucose) and further conversion to other by-products.



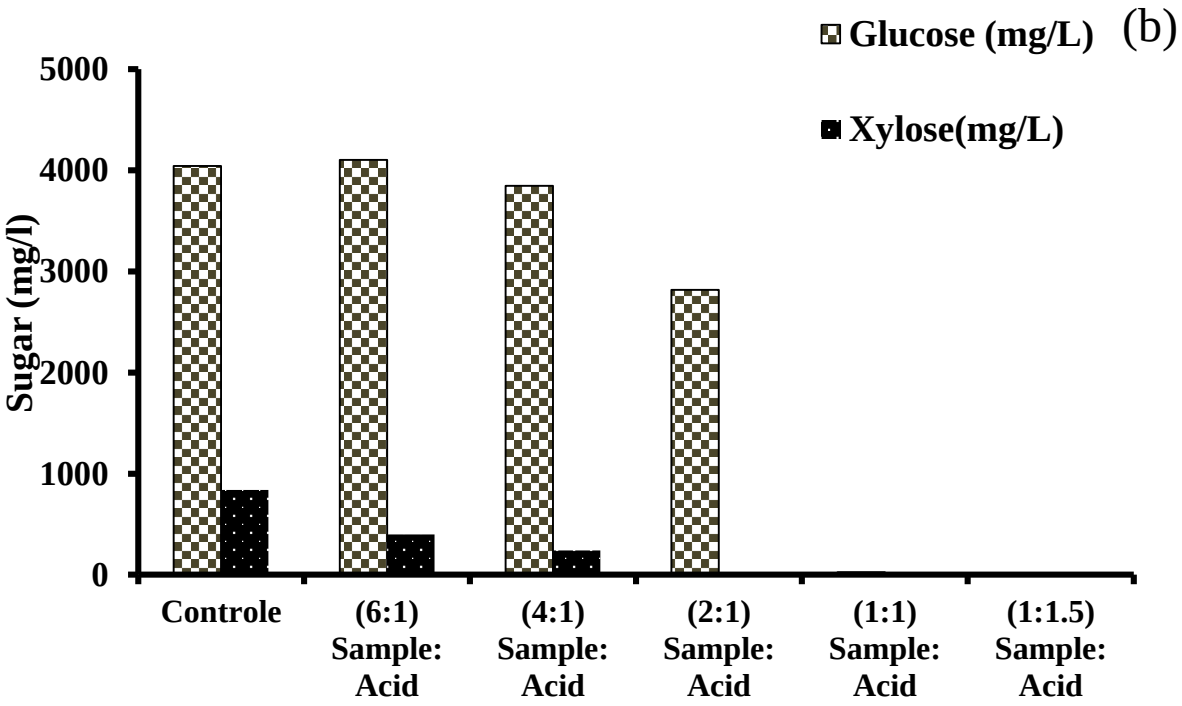
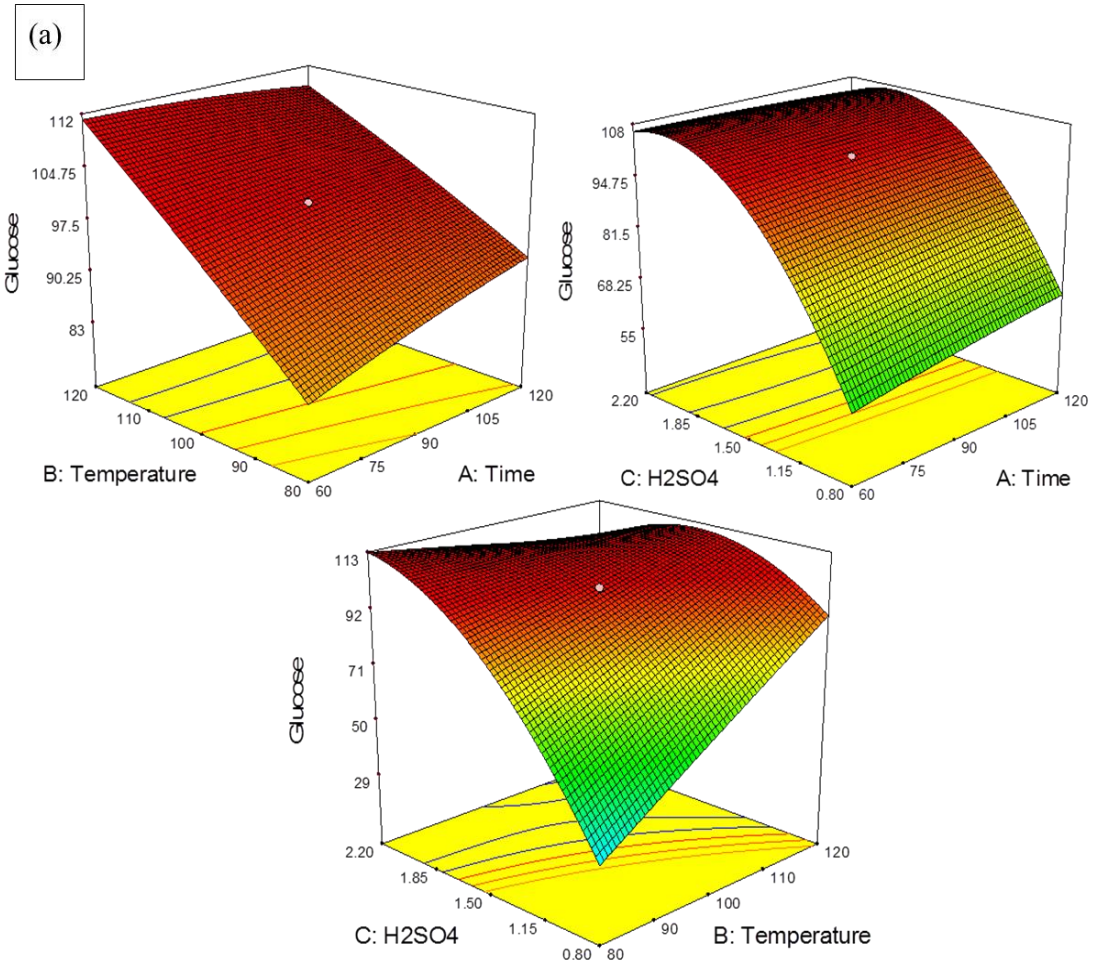


Figure-2 Effect on sugar removal from waste effluent of nano-crystalline cellulose (NCC) manufacturing process; (a): heating on in-situ hydrolysis; **(b)**: acid mediated heating



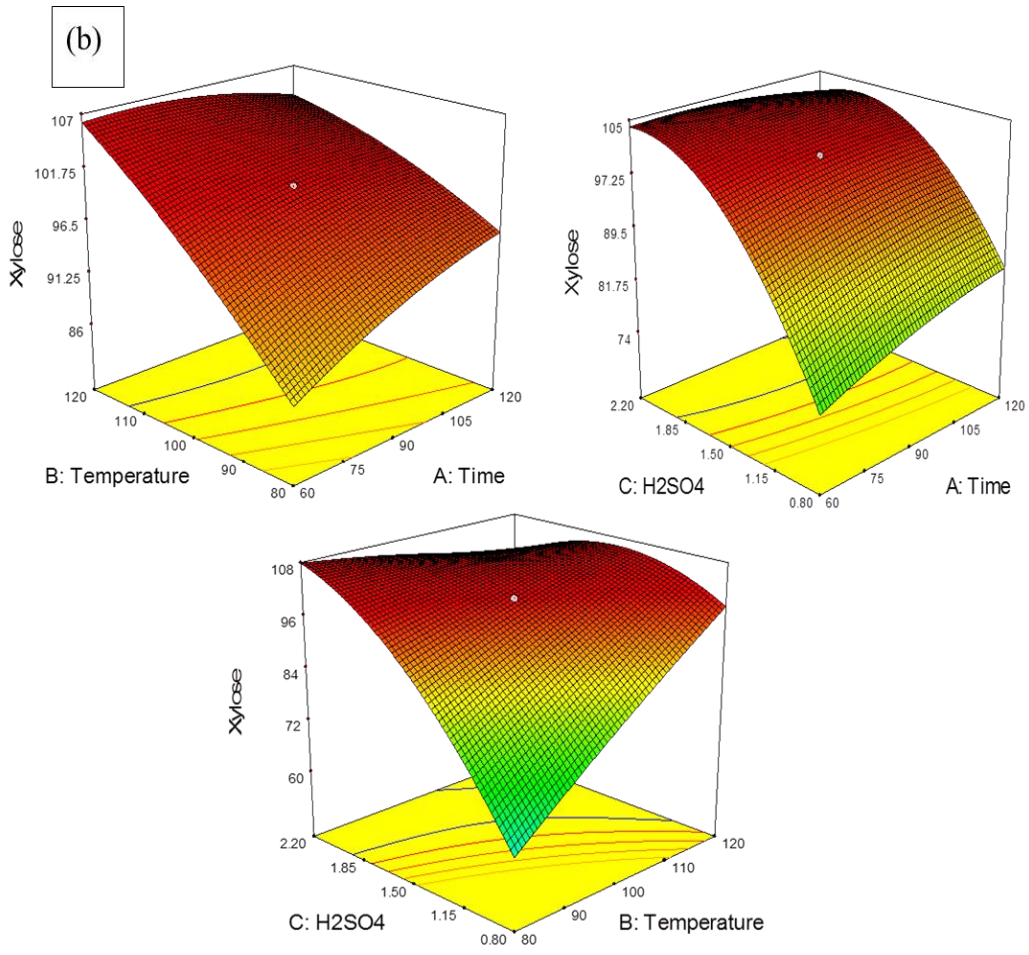


Figure-3 : Effect of different parameters on acid mediated heating to remove (a) glucose and (b) xylose from waste effluent from nano-crystalline cellulose (NCC) manufacturing processing

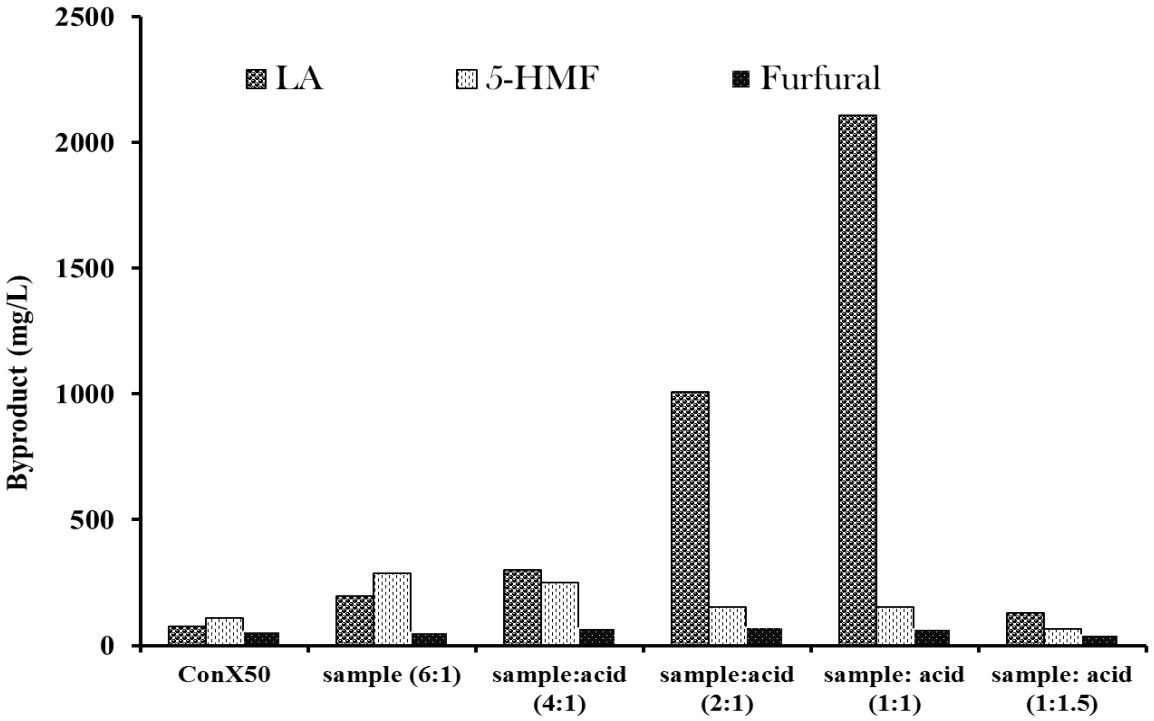


Figure-4: Effect of acid mediated heating on by-product formation to remove sugars from the waste effluent from nano-crystalline cellulose (NCC) manufacturing process.

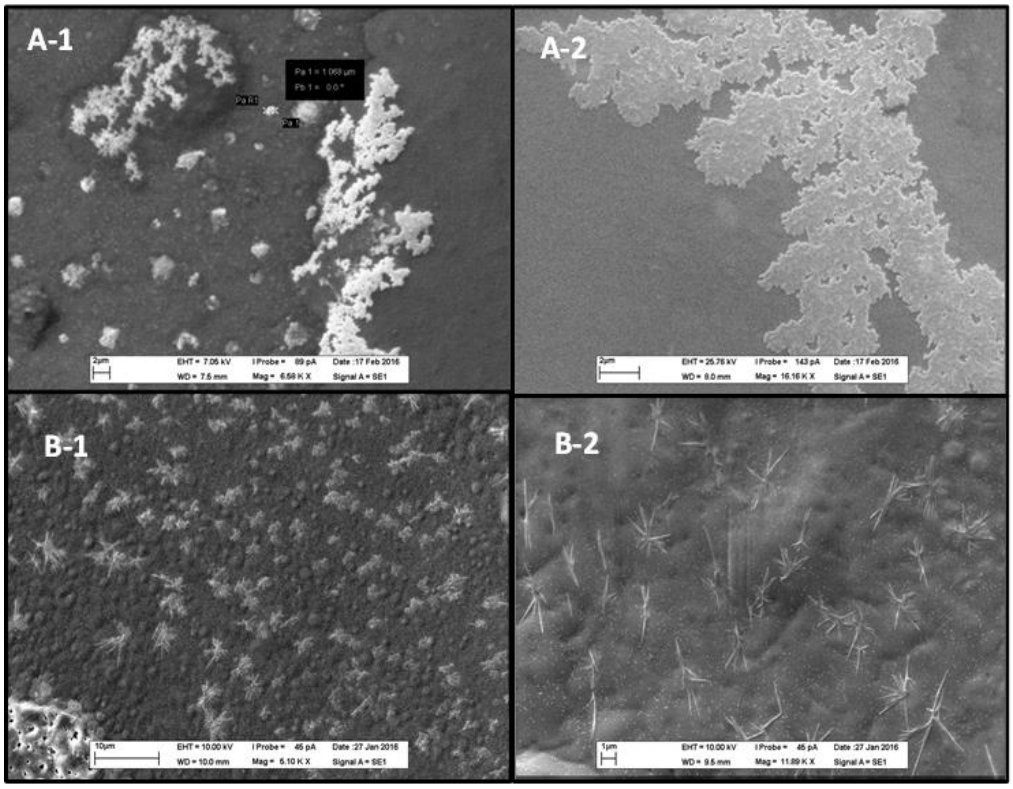


Figure-5: Scanning electron microscope image where A-1: waste sample without treatment at lower magnification; A-2: waste sample without treatment at higher magnification; B-1: waste sample with acid mediated heating treatment at lower magnification; B-2: waste sample with acid mediated heating treatment at higher magnification.

Table-1: Various factors and their limits considered for the study using response surface methodology

Serial Number	**Factor**	**Variable**		**Coded levels**			
			-α	Low	Middle	High	+α
1	A	Temperature	66.36	80	100	120	133.63
2	B	Time (min)	39.54	60	90	120	140.45
3	C	H_2SO_4	0.32	0.8	1.50	2.20	2.68

Table-2: Experimental design and the responses for removal of sugar from nano-crystalline cellulose processing acid effluent

Run	Factor 1	Factor 2	Factor 3	Response 1	Response 2	Response 3
	A:Time	B:Temperature	C:H_2SO_4	%Glucose removal	%Xylose removal	% Treholose removal
1	120.00	80.00	0.80	35.340	63.21	99.9
2	90.00	100.00	1.50	99.937	99.98	99.9
3	90.00	100.00	1.50	99.937	99.98	99.9
4	90.00	66.36	1.50	99.932	99.98	99.9
5	90.00	100.00	1.50	99.937	99.98	99.9
6	90.00	133.64	1.50	99.957	99.99	99.9
7	60.00	120.00	2.20	99.944	99.987	99.9

8	60.00	80.00	0.80	13.669	40.98	81.20
9	60.00	120.00	0.80	99.923	99.962	99.9
10	120.00	80.00	2.20	99.925	99.9618	99.9
11	39.55	100.00	1.50	99.917	99.9543	99.9
12	90.00	100.00	1.50	99.937	99.98	99.9
13	120.00	120.00	0.80	99.909	99.913	99.9
15	120.00	120.00	2.20	99.990	99.98	99.9
16	90.00	100.00	1.50	99.937	99.98	99.9
17	140.45	100.00	1.50	99.99	99.99	99.9
18	90.00	100.00	0.32	6.183	60.9	99.9
19	90.00	100.00	2.68	99.900	99.9	99.9
20	60.00	80.00	2.20	99.980	99.95	99.9

Table 3. Analysis of variance (ANOVA) for the fitted quadratic polynomial model for removal of sugar from nano-crystalline cellulose processing acid effluent.

Source	**Sum of squares**	**Degree of freedom**	**Mean square**	***p*-value***
Model	15869.50	9	1763.28	< 0.0004
A-Time	34.70	1	34.70	0.06457
B-Temperature	1667.20	1	1667.20	0.0082*
C-(Sample:H_2SO_4)	6973.75	1	6973.75	< 0.0001*
AB	58.25	1	58.25	0.5529
AC	58.68	1	58.68	0.5514
BC	2842.36	1	2842.36	0.0016*
A^2	3.26	1	3.26	0.8874
B^2	3.30	1	3.30	0.8867
C^2	4195.01	1	4195.01	0.0004*
Residual	1544.80	10	1415.86	-
Lack of fit	1544.80	5	2123.79	-
Core total	17414.30	19	- -	-

* Significant ($p < 0.05$)