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Original Research Article

Sonolytic, sonocatalytic and sonophotocatalytic degradation of carboxymethyl cellulose (CMC): kinetic and mechanisms

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ABSTRACT

In this research, enzymatic degradation of carboxymethyl cellulose (CMC) with different concentration of substrate was studied using stable available parameters of Michaelis-Menton and calculated maximum reaction rate V_{max}. We also used effective approaches such as ultrasonic, sonocatalytic and sonophotocatalytic irradiation in the presence of TiO_2 nanoparticles as pretreatment. Degradation of the cellulose by means of ultrasound irradiation and its combination with heterogeneous (TiO_2) was investigated. The emphasis was on evaluating the effect of additives on degradation rate constants. Ultrasound irradiation (24 kHz) was provided by a sonicator, while an ultraviolet source of 16 W was used for UV irradiation. The extent of sonolytic degradation increased when ultrasound power (in the range 1560 W) increased, and the presence of TiO_2 did not have significant effect on degradation. We should also note that TiO_2 sonophotocatalysis led to complete cellulose degradation in 120 min with increasing the catalyst loading. TiO_2 sonophotocatalysis was always faster than the respective individual processes due to the enhanced formation of reactive radicals as well as the possible ultrasound-induced increase of the active surface area of the catalyst. Their efficacy on enhancement of reactivity was discussed based on the kinetic parameters including, Michaelis constant K_m, maximum reaction rate V_{max} and initial reaction rate, correlating with ultrasonic conditions. Also, values of K_m and V_{max} were calculated in the absence and presence of ultrasonic and sonophoto waves.

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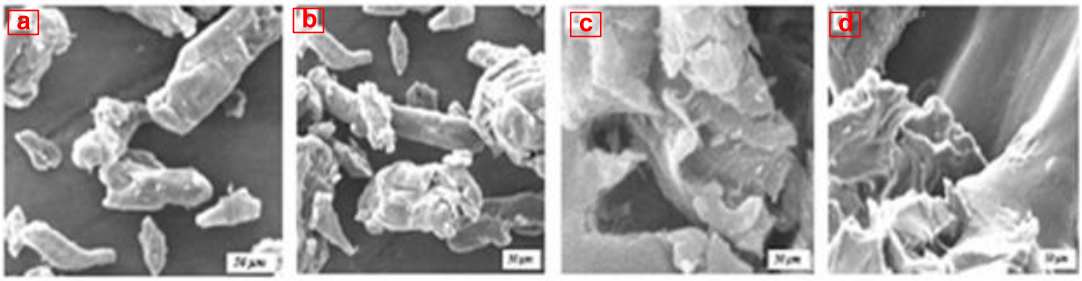
Graphical Abstract



a) CMC
b) CMC enzymatic degradation with sono process
c) CMC enzymatic degradation with photo process
d) CMC enzymatic degradation with sono-photo process

Introduction

The enzymatic process is more attractive because of environmental considerations. The conversion efficiency can be enhanced by adjusting the process conditions, or by enzyme genetic modification [1–4]. Cellulose is the most abundant carbohydrate component of biomass. Along with the diminishing of fossil fuel resources and global heating warnings caused by greenhouse gas emissions, the efficient utilization of cellulosic biomass is gaining more attention [5, 6]. Cellulose is a biopolymer of glucose. There are many methods to hydrolyze cellulose into glucose. Therefore, glucose as a platform molecule will play a significant role in future chemical and biochemical industries [7]. Cellulase is an important enzyme complex which can hydrolyze cellulose to form oligosaccharides and glucose. Cellulase usually consists of endoglucanase, cellobiohydrolase, and *b*-glucosidase which work synergistically to hydrolyze cellulose [8]. Celluloses also play an important role in other specialized commercial applications such as fabric medication's [9], paper and pulp industry [10], and food industry [11]. Recently, applications of low-frequency (1 MHz) and high-intensity (0.3 W/cm^{-2}) ultrasound techniques in laboratory and food industry have been at the center of attention in order to depolymerize macromolecules, make emulsions, disrupt biological cells, and deflocculated droplets. Degradability of polymers is a critical functionality of their applications. Currently, no official standard method has been established in the determined biodegradability of polymers. The enzyme method [12] the microbiological method [13] and the soil burial method [14, 15] have been used by different researchers. Moreover, the biodegradability was also recorded by diverse indexes even in the same method [16]. In this work, we aimed at reporting the availability of the three approaches to enhance the reactivity of cellulose hydrolysis using a pretreatment by

ultrasonic, sonocatalytic, and sonophotocatalytic irradiation in the presence of TiO_2 nanoparticles pretreatment.

Experimental

Materials and methods

Carboxymethyl cellulose (CMC) and hydroxide sodium provided by Fluka company. 3,5-dinitro salicylic acid, phenol, potassium sodium tartrate, calcium chloride, sodium sulfite (for preparing DNS reagent) and glucose (for preparing satandard curve) were provided by Merck company. Cellulase was provided by *Aspergillus* niger (specific activity; 111 U/mg^{-1}). Infrared spectra with a resolution of 4 cm^{-1} of the samples as KBr pellets were recorded by Shimaszu FT-IR RF50 spectrometer. Morphology of the cellulose before and after biodegradation was investigated using a scanning electron microscope (SEM, model XL30, Netherland). The substrates were covered with pure metallic Ag. The laying down of Ag was carried out using evaporation of the metal under a high vacuum to give a thickness of around 100 Å.

Pretreatment of substrate by ultra sonication

A desired amount of CMC was suspended in 0.1 M acetate buffer solution, pH 4.8, and the suspension was pretreated by ultrasonic irradiation at 323 K. Generally, for any polymer degradation process to meet the industry requirements, specification of the sonication conditions (appropriate irradiation power, irradiation frequency, temperature, concentration and irradiation time), which lead to a particular relative molar mass (RMM) distribution is of high importance. In this study, reactions were carried out in a cylindrical 100 mL pyrex glass vessel which is schematical. An ultrasound generator (Dr. Hielscher Ultrasonic Processor UP200 H) operating at a fix frequency of 24 kHz with a variable power output up to 100 W nominal value, in aqueous media was used for sonication experiments. A titanium-made H3 sonotrode (u=3 mm) immersed in liquid from the open to the atmosphere top of the vessel was used to deliver the ultrasound energy in the reaction mixture. The bottom of the vessel was fitted with a glass cylindrical tube housing the light source. There was a pair of 8 W, UV lamp which emits in the 200-300 nm wavelength range with a maximum at 254 nm. The vessel was fed with a 100 mL CMC solution and the reaction temperature in the case of sonolysis, sonocatalysis and sonophotocatalysis was kept constant at 50 ± 1 °C through the use of cooling water circulating through the double-walled compartment, thus acting as cooling jacket. The reaction vessel was covered with a dark cloth to avoid unwanted photochemical reactions induced by natural light. Different treatments were tested, namely: sonolysis (US), photocatalysis (UV), combined sonolysis

and photocatalysis (US + UV). For the experiments in the presence of TiO_2, a concentration range 0.1-0.5 g/L^{-1} of TiO_2 nanoparticles was used.

Enzymatic hydrolysis of pretreatment of substrate by ultra sonication

A cellulose suspension (2-10 g/L^{-1}) and an enzyme solution (0.5 g/L^{-1}) were prepared in 100 mL of 0.1 M acetate buffer solution, pH 4.8. Both were preheated to 50 °C. The reaction mixture was stirred magnetically at a high stirring speed to obtain well-reproducible data. The temperature was controlled at 323 °K throughout the reaction. The reducing sugars in the degradation solutions were quantified by the dinitrosalicylic acid (DNS) method: 1 mL of reagent DNS was added to 1 mL of the sample to be analyzed using 1 mg/mL glucose stock solution as a standard. At the same time, the blank was prepared using 1 mL of control sample. The mixture was heated at 388-398 °K for 10 min. After cooling at room temperature, 5 mL of distilled water was added. Samples were taken at desired times and centrifuged for 5 min. The glucose concentration in the supernatant was determined using DNS method. The absorbance was measured at 540 nm using a UV-vis spectrophotometer.

Results and Discussion

Enzymatic hydrolysis of cellulose without pretreatment

Results of our experiments revealed that, concentration of the glucose curve was drawn during 120 min. Thereby; the production of glucose was used as a measure of cellulose hydrolysis. The extent of glucose over a 120 min hydrolysis time for each substrate is shown in Figures 1 and 2. This illustrates the initial rate of glucose production by each substrate up to a hydrolysis time 10 min. Moreover, a linear relationship between the concentration of glucose and time for the first 10 min of hydrolysis has also been presented.

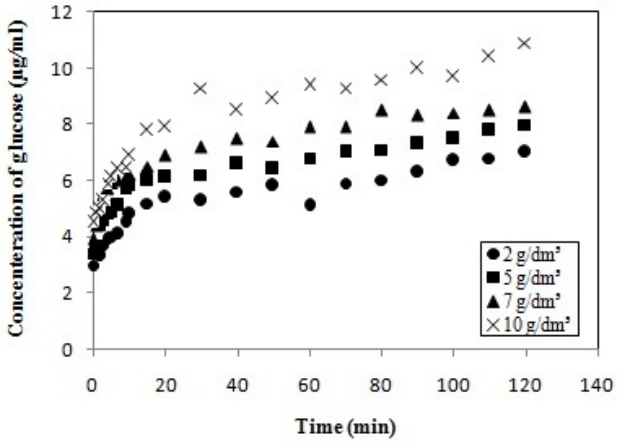


Figure 1. Glucose production from hydrolysis of CMC due to the enzymatic attack by cellulase for 120 min without ultrasonication pretreatment. $[S]_0$ = 2-10 g/L^{-1} buffer; $[E]_0$ = 0.5 g/L^{-1} buffer

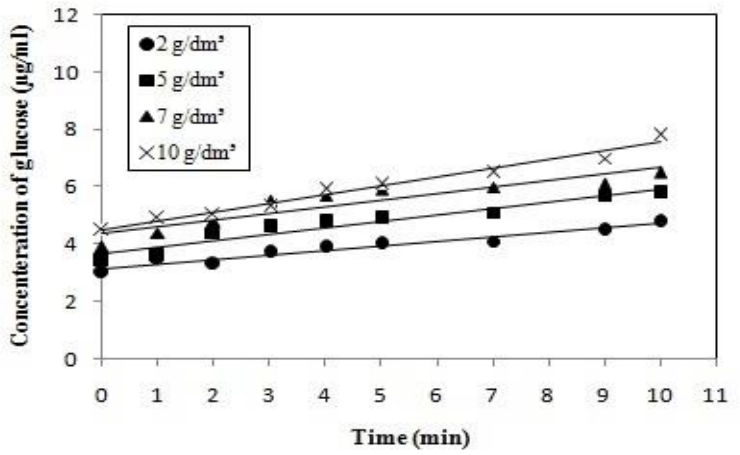


Figure 2. Glucose production from hydrolysis of CMC in the first 10 min of enzymatic degradation due to the action of Cellulase without ultrasonication pretreatment. $[S]_0$ = 2-10 g/L^{-1} buffer; $[E]_0$ = 0.5 g/L^{-1} buffer

Speed versus (vs) time curves until first 10 min that it is speed reaction is first degree and after 10 min, by reason effect of products determination, inactivate enzyme and decreasing level substrate reaction able, speed of reaction decreases and remains constant. In this section, the speed of reactions is zero. Gradient of concentration of glucose vs. time curves is in first 10 min of reaction (that it is straight line). This cruve will give first speeds. Although the cellulose used in this paper was optically suspended in the reaction solution under each reaction condition, the experimental results were well consistent and highly reproducible, indicating that it was sufficient to obtain reliable data for the analysis of the reaction kinetics. Figure 1 reveals typical time courses of the enzymatic hydrolysis of cellulose, without ultrasonication pretreatment. Hydrolysis reaction began immediately after addition of the enzyme solution, and then glucose production was leveled off.

Figure 2 depicts the curve first 10 min of enzymatic hydrolysis due to the action of cellulase is straight line.

Effect of the pretreatment on the hydrolysis

Improving the hydrolysis rate before the enzyme reaction causes CMC in our solution to be pretreated by sono, sonocatalytic and sonophotocatalytic pretreatments in the presence of TiO_2 nanoparticles. The kinetic parameters for different pretreatments are shown in Table 1.

The ultrasonic irradiation time is mostly effective within the 30 min irradiation time to enhance the initial reaction rate V_{max}. When using ultrasonic pretreatment to substrate the hydrolysis of CMC recourses, we recommend the conditions of high irradiation power and short irradiation time for increasing degradation rate under the same irradiation energy conditions. It was hardly influenced

by the ultrasonic irradiation. The efficacy of the ultrasonic irradiation in pretreatment on reaction rate was demonstrated based on our original kinetic data. In addition, the effect of the ultrasonic irradiation time in pretreatment on V_{max} was examined at 30 W of irradiation power (P_{ir}). The results in Figure 3 shows the effect of increasing changes in ultrasound power on the concentration of glucose (μg/mL^{-1}) of CMC solution as a function of the sonication time at 5 g/L^{-1} initial polysaccharide concentration under air.

Table 1. Kinetic parameters for different substrates

Sub.	V_{max} (g/L^{-1}/s^{-1})	K_m (g/L^{-1})
No irradiation	11.103	17.4
US	14.705	17.91
US+0.1 g/TiO_2	15.733	18.93
US+0.2 g/TiO_2	16.404	18.9
US+0.3 g/TiO_2	16.198	19.48
US+0.4 g/TiO_2	17.235	19.89
US+0.5 g/TiO_2	17.289	20.43
UV+0.1 g/TiO_2	12.434	17.56
UV+0.2 g/TiO_2	13.653	18.01
UV+0.3 g/TiO_2	13.896	18.34
UV+0.4 g/TiO_2	14.008	18.48
UV+0.5 g/TiO_2	14.87	18.42
US+UV	15.898	20.34
US+UV+0.1 g/TiO_2	17.805	23.87
US+UV+0.2 g/TiO_2	19.089	24.81
US+UV+0.3 g/TiO_2	20.569	26.98
US+UV+0.4 g/TiO_2	21.901	27.86
US+UV+0.5 g/TiO_2	23.875	29.45

According to our observation, the concentration of glucose (μg/mL^{-1}) increased with increasing the nominal applied power from 15 to 60 W. The Effect of the presence of catalyst TiO_2 and catalyst concentration in constant power of ultrasound (30 W) on the degradation rates was also investigated. Figure 4 illustrates a change in the glucose concentration (μg/mL^{-1}) which is in complete contrast to the sonication time in the sonocatalytic process (US+TiO_2).

Figure 5 depicts the changes in the glucose concentration (μg/mL^{-1}) versus sonication time in the sonocatalytic process (US+TiO_2).

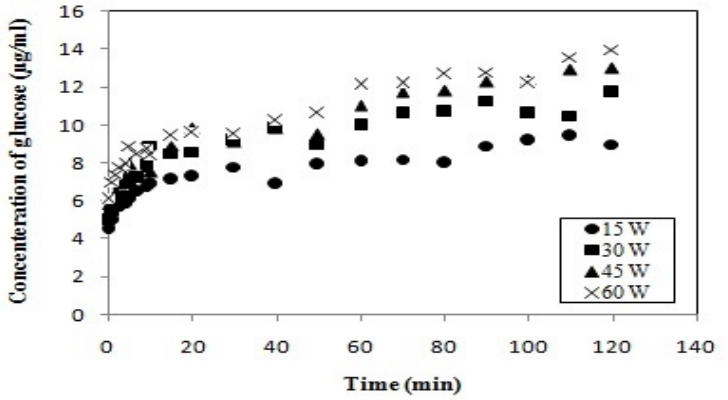


Figure 3. Effects of ultrasonication for glucose production from hydrolysis of CMC $[S]_0$ = 2 g/L^{-1} buffer; $[E]_0$ = 0.5 g/L^{-1} buffer; irradiation time = 30 min; P_{ir} = 15-60 W

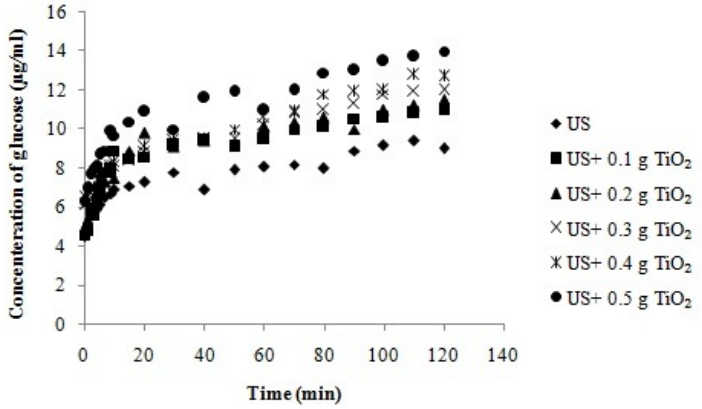


Figure 4. The plot of concentration of glucose (µg/mL^{-1}) versus the time in sonocatalytic process, for different loading of catalyst at constant power of ultrasound (30 W). $[S]_0$ = 2 g/L^{-1} buffer; $[E]_0$ = 0.5 g/L^{-1} buffer

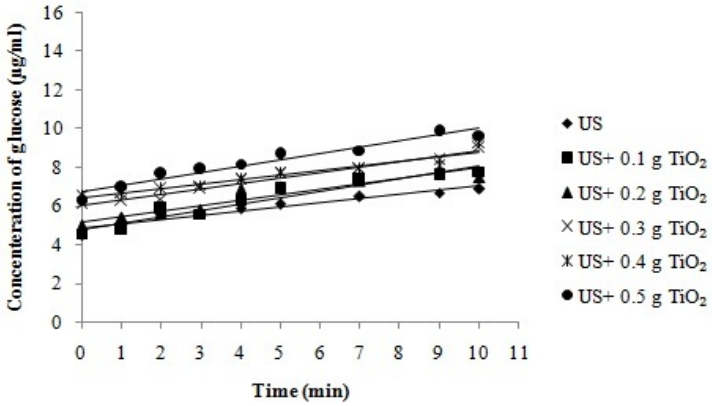


Figure 5. The plot of concentration of glucose (µg/mL^{-1}) versus the time in sonocatalytic process, for different loading of catalyst at constant power of ultrasound (30 W). $[S]_0$ = 2 g/L^{-1} buffer; $[E]_0$ = 0.5 g/L^{-1} buffer

Figure 6 shows a change in concentration of glucose (μg/mL^{-1}) versus sonication time in the sonocatalytic process (UV+TiO_2).

Figure 7 illustrates the changes in concentration of glucose (μg/mL^{-1}) versus sonication time in the sonocatalytic process (US+UV+TiO_2). In general, particles may enhance degradation providing additional nuclei for bubble formation. However, an imperfect effect may also occur because of sound attenuation. As seen, the presence of TiO_2 particles in the reaction mixture caused a partial increase in the sonochemical degradation of hydrolysis of cellulose.

The reaction system in this research is homogeneous one and the homogeneous systems show good following from Michaelis–Menten. The Michaelis constant (K_m) and the maximum reaction rate (V_{max}) for glucose production in the reactions were calculated and are given in Table 1. Effect of ultrasonic waves at the speed of the starch enzyme hydrolysis value of glucose production was measured with time under ultrasonic waves. the aim of this research is to show that when polymer, before enzyme hydrolysis impress of ultrasonic waves, cuts and the enzyme can place on substrate easily, and abstract effects of spatial decreases. In fact, more surface of substrate is in control of enzyme and the field of enzyme activity for enzyme becomes wider.

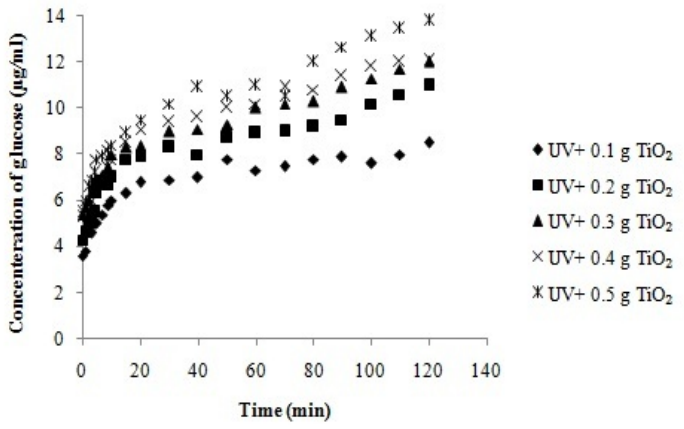


Figure 6. The relationship between concentration of glucose (μg/mL) and enzymatic degradation time in photocatalytic process, for different loading of catalyst at constant power of ultraviolet (16 W). $[S]_0$ = 2 g/L^{-1} buffer; $[E]_0$ = 0.5 g/L^{-1} buffer

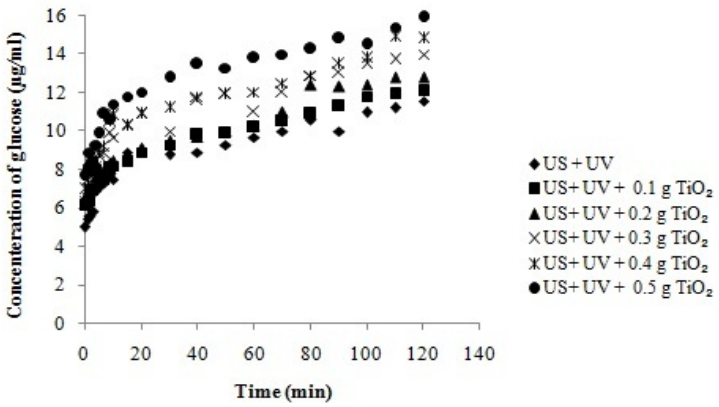


Figure 7. The relationship between concentration of glucose (μg/mL) and enzymatic degradation time in sonophotocatalytic process, for different loading of catalyst at constant power of ultrasound (30 W) and ultraviolet (16 W). $[S]_0$ = 2 g/L^{-1} buffer; $[E]_0$ = 0.5 g/L^{-1} buffer

FT-IR spectra

The FT-IR spectrum of undegraded and degraded blend in the presence of cellulose is shown in Figure 8.

As mentioned before, this study focused on the range of the absorption pattern in six main regions: (a) stretching vibration of the O-H group between 3436 and 3455 cm^{-1}; (b) Stretching vibration of the C=O at about 1740 cm^{-1}; (c) Stretching vibration of the C=C at about 1638 cm^{-1}; (d) Stretching vibration of the C-O at about 1237 cm^{-1}; (e) Stretching vibration of the C-H at about 1454 cm^{-1}; (f) Stretching vibration of the COO^- group between 1500-1600 cm^{-1}. The absorbance at 3400-3500 and 1060 cm^{-1} are more sensitive to the conformational changes produced during degradation processes, thus one can surmise that it indicates a short range order and helicity changes when crystallinity and molecular orientation are lost. After degradation with enzymes, the intensity of the peak at 1500 and 1600 cm^{-1} decreased, which shows the action of cellulase in cleaving the glycoside linkages of cellulose. The absence of the peak in the 1740-1720 cm^{-1} range indicates the absence of aldehyde groups from which it is inferred that all aldehyde groups were involved in crosslinking.

Scanning electronic microscopy (SEM)

Several SEM images of CMC are demonstrated in Figure 9. We can see that the CMC in sono photocatalytic process completely degraded.

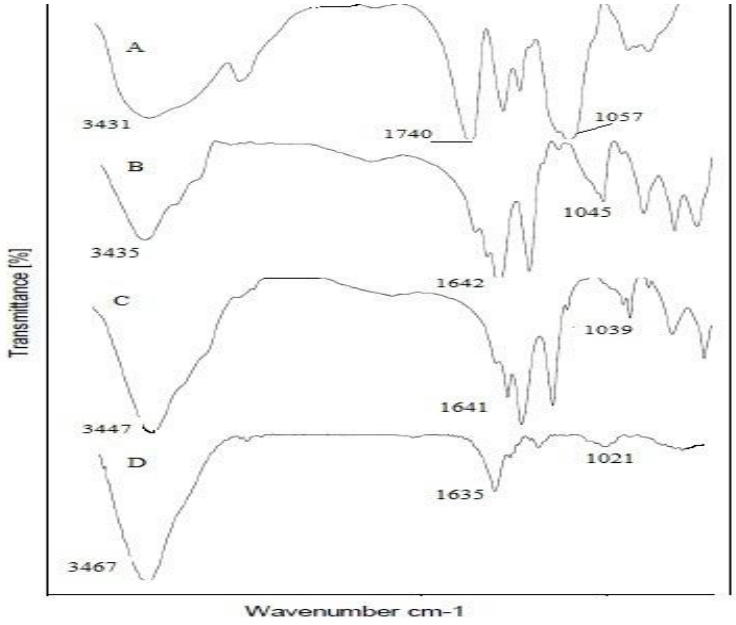


Figure 8. FT-IR spectra of cellulose: a) CMC degraded; b) CMC enzymatic degradation time in sonocatalytic process degraded; c) CMC enzymatic degradation time in photocatalytic process degraded; d) CMC enzymatic degradation time in sonophotocatalytic process degraded

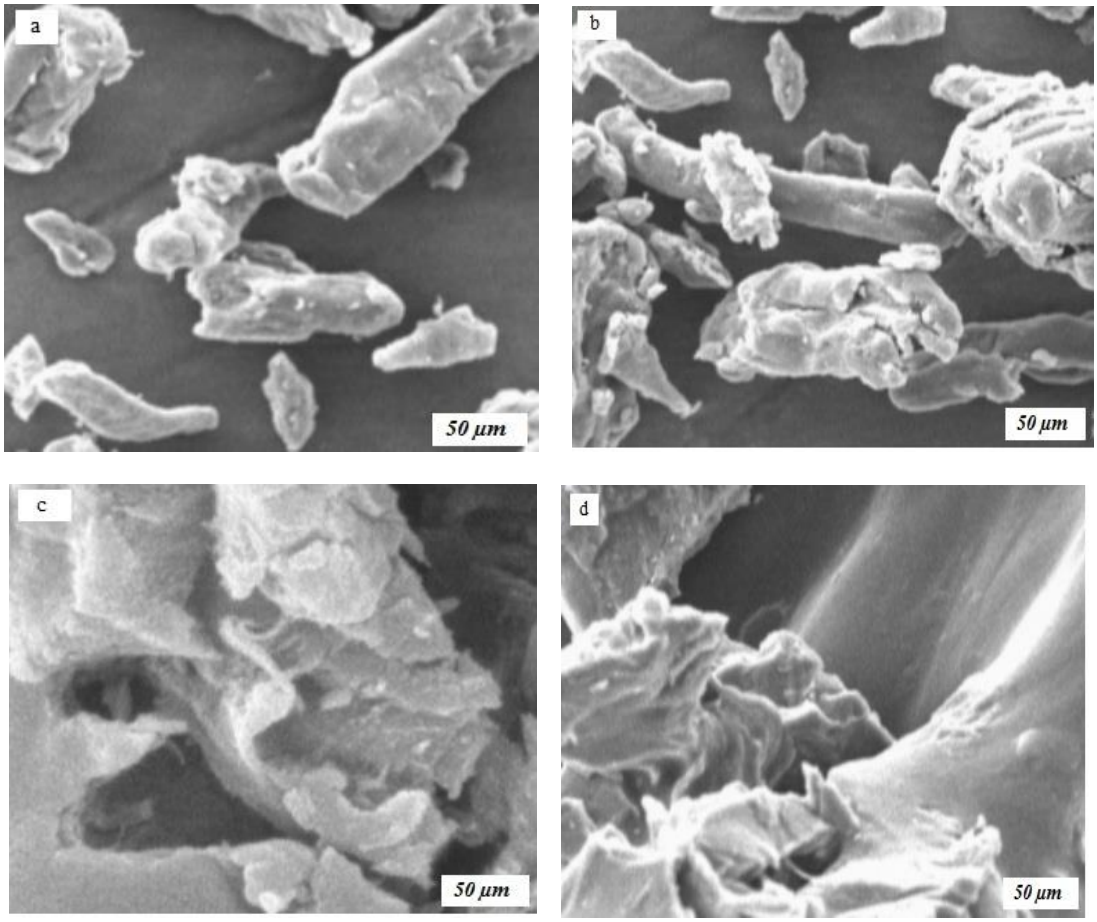


Figure 9. Scanning electron micrographs of CMC degradable in 120 min of enzymatic degradation due to the action of cellulase: a) CMC degraded; b) CMC enzymatic degradation time in sonocatalytic process degraded; c) CMC enzymatic degradation time in photocatalytic process degraded; d) CMC enzymatic degradation time in sonophotocatalytic process degraded

Conclusions

In this research, we observed that enzymatic hydrolysis of CMC with increasing concentration of substrate increased the value of glucose production and hydrolysis speed, whereas after 10 min because of the interference of different factor of high concentration of substrate, speed became stable with zero degrees. This experiment tried at developing a high-performance system for the enzymatic hydrolysis of CMC. Ultrasonic waves of CMC hydrolytic are reactivity over without ultrasonic waves. Kinetic analysis revealed that a 30 min of irradiation and 30 W Pir was optimal in enhancing V_{max}. Pretreatment of starch in suspension by ultrasonic irradiation further developed the reaction rate. Enhancement of V_{max} by ultrasonication was increased around 5 times that received without ultrasonication pretreatment. On the other hand, the Km in the ultrasonically treated system remained at almost the same level as that in the untreated system. Finally, increasing the irradiation

power could markedly reduce the time needed for ultrasonication. These results are promising for the development of a practical system for the scarification of CMC resources.

Disclosure Statement

No potential conflict of interest was reported by the authors.

References

[1]. Bajpai A.K., Shrivastava J. *Polym Int.,* 2005, **54**:1524

[2]. Lv D., Xu M., Liu X., Zhan Z., Li Z., Yao H. *Fuel Proc. Technol*., 2010, **91**:903

[3]. Sutarlie L., Yang K.L. *J. Coll. Interface Sci*., 2013, **411**:76

[4]. Kim M.N., Lee A.R., Yoon J.S., Chin I.J. *Eur. Polym. J.,* 2000, **36**:1677

[5]. Rutkowski P. *Fuel Proc. Technol*., 2011, **92**:517

[6]. Xu Y.X., Hanna M.A. *Carbohydr. Polym.,* 2005, **59**:521

[7]. Demirgoz D., Elvirs C., Mano J.F., Cunha A.M., Piskin E., Reis R.L. *Polym. Degrad. Stab*., 2000, **70**:161

[8]. Galbe M., Zacchi G. *Biofuels*., 2007, **41**:65

[9]. Himmel M.E., Ding S.Y., Johnson D.K., Adney W.S., Nimlos M.R., Brady J.W., Foust D. *Science*., 2007, **315**:804

[10]. Kumar P., Barrett D.M., Delwiche M.J., Stroeve P. *Ind. Eng. Chem. Res*., 2009, **48**:3713

[11]. Zhang Y.H., Lynd L.R. *Biotechnol. Bioeng.*, 2004, **88**:797

[12]. Benedict C.V., Cook W.J., Jarrett P., Cameron J.A., Huang S.J., Bell J.P. *J. Appl. Polym. Sci*., 1983, **28**:327

[13]. Lavenson D.M., Tozzi E.J., Karuna N., Jeoh T., Powell R.L., McCarthy M.J. *Bioresour. Technol*., 2012, **111**:240

[14]. Xiao C., Yang M. *Polym,* 2006, **64**:37

[15]. Vaezifar S., Faghihian H., Kamali M. *Iran J. Chem. Chem. Eng*., 2009, **28**:23

[16]. Tavasoli A., Irani M., Nakhaeipour A., Mortazavi Y., Khodadadi A.A., Dalai A.K. *Iran. J. Chem. Chem. Eng*., 2009, **28**:37

[17]. Imhoff M.L., Bounoua L., Rickett T., Loucks C., Harriss R., Lawrence W. *Nature*., 2004, **429**:870

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