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Enhanced biogas production from sunflower stalks using hydrothermal and organosolv pretreatment

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ABSTRACT

Biomethane production through anaerobic digestion of sunflower stalks was improved by the hydrothermal and isopropanol-based organosolv pretreatments. The pretreatments were conducted at different temperatures (140, 160, 180, and 200 °C) for 30 and 60 min with/without addition of 1% sulfuric acid. The pretreatment of stalks with 50% (v/v) aqueous isopropanol containing 1% w/w (based on dry stalks) sulfuric acid at 160 °C resulted in the highest lignin removal. Methane production yield of the pretreated substrate was improved by 45–124% compare to 124 mL CH_4/g VS obtained from the digestion of untreated stalks. In the best case, hydrothermal pretreatment at 180 °C for 60 min and organosolv pretreatment at 160 °C for 30 min with 1% H_2SO_4 followed by 45 days anaerobic digestion resulted in 234 and 278 mL CH_4/g VS, respectively. Structural analyses of the stalks indicated that lignin removal, crystallinity reduction, and structural modifications by the pretreatments were the main reasons for the improved biogas production.

1. Introduction

Biomethane, a renewable source of energy, is currently considered as a respectable alternative to replace fossil fuels for heating, electricity production, transportation fuel and production of value-added chemicals (Teghammar et al., 2012; Weiland, 2010). All types of raw materials containing carbohydrates, fats, proteins, cellulose, and hemicelluloses as the major components can be employed as substrates for biomethane production (Weiland, 2010). In particular, lignocellulosic materials such as agricultural residues have been widely considered as the most suitable raw materials for biomethane production due to relatively low costs, high availability, and no direct competition with food and feed production (Shafiei et al., 2013; Taherzadeh and Karimi, 2008). Sunflower residues, especially sunflower stalks which are lignocellulosic biomass, are globally produced 78–182 million tons per year (reported by FAO in 2013) and typically disposed as a waste or burned in the fields causing environmental pollution (Ruiz et al., 2013). In contrast, high cellulose and hemicellulose contents of sunflower stalk makes it a potential feedstock for biomethane production (Ziebell et al., 2013). However, the highly crystalline cellulose is well packed by a matrix of lignin and hemicellulose in the natural structure, and it is the main drawback of using lignocellulosic materials limiting the production of methane (Frigon and Guiot, 2010; Taherzadeh and Karimi, 2008). Lignin, as a main factor of integrity and structural rigidity, is responsible for the recalcitrance of lignocellulosic substrate to bacterial degradation by limiting cellulose accessibility (Gupta et al., 2011; Shafiei et al., 2013). Therefore, an effective pretreatment step is required to enhance the digestibility of lignocellulose (Bateni et al., 2014; Hendriks and Zeeman, 2009). Alkaline and oxidative pretreatments have been evaluated for biomethane production from sunflower stalks (Monlau et al., 2012). Although these processes are often performed at a low temperature, they are associated with a high energy consumption and high wastewater production.

Hydrothermal pretreatment by hot water has been applied for the pretreatment of lignocellulosic biomass over last few decades (Taherzadeh and Karimi, 2008). In this process, water penetration into the biomass structure increases the accessible and susceptible surface area of cellulose by removing hemicellulose and a part of lignin (Chandra et al., 2012). Organosolv pretreatment is about the same as hydrothermal pretreatment; however, the lignin and hemicellulose removals are conducted using organic liquid solvents such as alcohols, ketones, glycols, phenols, and ethers. This organic mixture partially hydrolyzes internal lignin and hemicellulose bonds, and consequently removes a considerable portion of lignin

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from the biomass. This phenomenon leads to an increase in the pore volume and surface area which can facilitate enzyme accessibility (Zhao et al., 2009). The recovery of relatively pure lignin as a value-added byproduct is a major advantage of organosolv pretreatment compared to other chemical pretreatments (Huijgen et al., 2012; Zhao et al., 2009). Lignin removal in a separate phase can also decline the economic and environmental problems associated with wastewater treatment of typical chemical pretreatment methods (Amiri et al., 2014). Furthermore, a strong inorganic catalyst, e.g., sulfuric acid, may be used to decrease the operating temperature or improve the delignification process (Zhao et al., 2009). These advantages along with the solvent recovery with minimal energy consumption have made organosolv process as one of the most promising methods to improve the conversion yield of different lignocellulosic materials for biofuel production (Amiri et al., 2014; Mesa et al., 2011; Wildschut et al., 2013; Zhao et al., 2009).

Alcohols, especially the low boiling point alcohols such as methanol and ethanol, appear to be the most frequent organic solvents used in alcohol-based organosolv pretreatment, due to their low cost and easy recovery (Zhao et al., 2009). However, methanol is a toxic chemical and forms flammable vapors at relatively low temperatures making the pretreatment process more complicated (Zhao et al., 2009). On the other hand, even though ethanol is less toxic than methanol, very high pressure is needed due to low boiling point of ethanol resulting in high equipment costs with accompanying safety concerns and maintenance difficulties (Zhao et al., 2009). Isopropanol is the other low boiling point alcohol which is similar to methanol and ethanol in solvent properties and evaporation rate. Unlike methanol and ethanol, isopropanol can be separated from aqueous solutions either through an evaporation step followed by condensation or by addition of a salt such as sodium chloride, sodium sulfate, or other inorganic salts (Othmer, 1999). Therefore, isopropanol can be employed for organosolv pretreatment of lignocellulosic biomass. However, based on our knowledge, there is no report in the literature on the utilization of isopropanol as a lignin solvent in the organosolv pretreatment of lignocellulosic materials for biofuel production.

The overall objective of this study is to evaluate the effect of hydrothermal and isopropanol organosolv pretreatment variables (pretreatment time and temperature) on the biomethane production from sunflower stalks. The effect of using sulfuric acid as a catalyst in both pretreatment processes on the lignin removal and biomethane production yields was also investigated. Moreover, the changes in the biomass structure and the reasons for the improvement were followed.

2. Materials and methods

2.1. Raw materials

Sunflower stalks were collected from a local field in the north of Isfahan (Isfahan, Iran) and dried in sunlight for 3 days. Stalks were milled and screened to achieve a size of less than 1 mm. The substrate was then dried at 105 °C in a convection oven to measure its dry weight content.

2.2. Organosolv and hydrothermal pretreatment procedures

Pretreatment experiments were carried out in a high pressure batch reactor with a working volume of 500 mL (Amiri et al., 2010). The reactor was loaded with 20 g (dry weight) of substrate and 200 mL of a 50% (V/V) isopropanol in water mixture (solid–liquid ratio of 1:10 (w/v)). In all pretreatment experiments, the ratio of biomass to organic solvent was kept constant the same as above and temperature and time of pretreatment were changed as below. The reactor was heated at a rate of 3 °C/min to a desired temperature (140, 160, 180, and 200 °C). The reactor was then kept for 30 or 60 min at the desired temperature to treat the biomass. Another set of pretreatments was conducted by adding 1% w/w sulfuric acid (based on dried weight of the stalk) as a catalyst. After the reaction time, the reactor was transferred to an ice bath. The solid phase was separated using filter, and then rinsed with 60 °C deionized water for several times until pH 7 and dried at room temperature. Finally, the pretreated samples were stored in resealable plastic bags at 4 °C until use.

Hydrothermal pretreatment was also performed under the same conditions as the organosolv pretreatment, except that the reactor was loaded with 200 mL water and 20 g substrate (dry weight) to obtain solid–liquid ratio of 1:10 (w/v).

2.3. Biogas production

The inoculum was obtained from a 7500 m^3 anaerobic digester (Isfahan municipal wastewater treatment plant, Isfahan, Iran) operating at 37 °C. Biogas production from the untreated and pretreated samples were performed in 118 mL serum glass bottles as anaerobic digesters at 37 °C (Hansen et al., 2004). Bottles were loaded with 20 mL inoculum and 0.25 g treated or untreated biomass (dry weight). By adding deionized water to each bottle, final volume was reached to 25 mL. Moreover, a sample containing 20 mL inoculum and 5 mL deionized water was used as a blank to be able to measure the gas production of inoculum alone. Bottles were capped by aluminum caps on butyl rubber stoppers. Then, to provide anaerobic conditions, bottles were purged using N_2 gas for about 2 min. The bottles were manually shaken every day and kept at 37 °C in an incubator. Gas samples were taken from the headspace of each bioreactor and analyzed using gas chromatography every three days during the first fifteen days and then every five days until the end of the experiment. All the experiments were carried out in duplicate. The biogas production results were analyzed using ANOVA toolbox of Microsoft Office Excel 2013. 2-factors ANOVA was used and the *p*-values less than 0.05 were considered as statistically significant.

2.4. Analytical methods

The composition of gas produced in the bottles (methane and carbon dioxide) was determined by a gas chromatograph (Sp-3420A, TCD detector, Beijing BeifenRuili Analytical Instrument CO.) equipped with a packed column (Porapack Q column, Chrompack). The carrier gas was nitrogen with flow rate of 45 mL/min. Temperatures of the injector, column, and detector were adjusted on 100, 40, and 150 °C, respectively. Carbohydrates, lignin contents, total solids, and volatile solids of treated and untreated sunflower stalks were analyzed according to the methods of Sluiter et al. (2008a,b). Sugars were analyzed by a high performance liquid chromatography equipped with RI and UV/vis detectors (Jasco International Co., Tokyo, Japan) and an Aminex HPX-87P column at 85 °C with deionized water as an eluent with a flow rate of 0.6 mL/min. Morphological changes in the treated and untreated sunflower stalks during the organosolv and hydrothermal pretreatments were observed by scanning electron microscopy (SEM). The dried samples were coated with a thin layer of gold and analyzed by a scanning electron microscope (KYKY-EM3200) at 26 kV.

The crystallinity and molecular structure of treated and untreated sunflower stalks were analyzed using a Fourier transform infrared (FTIR) spectrometer (Bruker Tensor 27 FTIR). The spectra were obtained with an average of 60 scans with 2 cm^{-1} resolution from 600 to 4000 cm^{-1}.

Table 1
Chemical composition and solid recovery of hydrothermal pretreated sunflower stalks.

Pretreatment conditions				Composition			
T (°C)	Time (min)	H_2SO_4 (%)	Glucan (%)	Xylan (%)	Arabinan (%)	Klason lignin (%)	Solid recovery (%)
140	30	–	38.6 ± 0.4	22.3 ± 0.4	1.4 ± 0.2	33.0 ± 0.6	73.6 ± 0.3
140	60	–	40.8 ± 0.7	21.9 ± 0.6	1.2 ± 0.3	32.1 ± 0.4	71.4 ± 0.5
140	30	1	40.7 ± 0.5	20.9 ± 0.5	1.0 ± 0.2	33.9 ± 0.7	71.1 ± 0.3
140	60	1	41.9 ± 0.3	20.4 ± 0.5	0.8 ± 0.3	33.5 ± 0.3	69.2 ± 0.7
160	30	–	43.4 ± 0.5	20.5 ± 0.6	1.0 ± 0.2	31.4 ± 0.4	70.6 ± 0.5
160	60	–	45.7 ± 0.7	19.4 ± 0.4	0.7 ± 0.2	30.6 ± 0.5	68.9 ± 0.2
160	30	1	45.6 ± 0.6	18.1 ± 0.5	0.6 ± 0.1	32.0 ± 0.4	67 ± 0.8
160	60	1	47.9 ± 0.4	17.4 ± 0.4	0.5 ± 0.2	31.4 ± 0.6	65.3 ± 0.4
180	30	–	49.7 ± 0.8	18.9 ± 0.7	0.5 ± 0.1	27.1 ± 0.8	67.8 ± 0.4
180	60	–	50.9 ± 0.6	18.0 ± 0.3	0.2 ± 0.1	26.7 ± 0.3	66.6 ± 0.6
180	30	1	53.1 ± 0.4	16.5 ± 0.7	0.2 ± 0.1	26.6 ± 0.7	63.1 ± 0.3
180	60	1	54.9 ± 0.5	16.0 ± 0.5	–	26.1 ± 0.3	61.2 ± 0.7
200	30	–	58.0 ± 0.6	11.0 ± 0.6	–	27.1 ± 0.3	
200	60	–	61.0 ± 0.3	9.1 ± 0.3	–	27.0 ± 0.4	
200	30	1	61.0 ± 0.5	9.0 ± 0.4	–	26.3 ± 0.3	
200	60	1	62.0 ± 0.4	7.0 ± 0.5	–	26.2 ± 0.4	

3. Results and discussion

3.1. Characterization of raw material

The untreated sunflower stalks, used in this study, consisted of (% dry basis): 34.1 ± 0.5 glucan, 24.4 ± 0.4 xylan, 1.8 ± 0.2 arabinan, and 26.8 ± 0.4 Klason lignin. Lignin contents of four accessions of cultivated sunflower have been reported within the range of 24.0–34.6% (Ziebell et al., 2013). The carbohydrate contents were similar to those reported by Caparrós et al. (2008) and Ruiz et al. (2013), although lignin content of the sunflower stalks used in this study was much higher than that of their reports. Dissimilarities in the composition may be partially attributed to dissimilarities in growing location, season, harvesting methods, and analysis methods.

3.2. Effects of pretreatment on the sunflower stalks composition and structure

Sunflower stalks were subjected to hydrothermal and isopropanol organosolv pretreatment with or without addition of sulfuric acid as a catalyst prior to anaerobic digestion in order to improve biogas production yield. The chemical composition of the hydrothermal and organosolv pretreated materials is presented in Tables 1 and 2, respectively. According to Table 1, total carbohydrate content of stalks was increased by all the pretreatments. The highest total carbohydrate content of hydrothermal pretreated stalks was 70.1%, achieved by pretreatment at 200 °C for 60 min (Table 1). Moreover, a minor increase (1.1%) in total carbohydrate content by hydrothermal pretreatment at lower temperature, i.e., 180 °C for 60 min, were achieved as a result of adding 1% w/w (based on dry stalk) H_2SO_4, compared to the hydrothermal pretreatment at 200 °C and the same retention time without H_2SO_4 (Table 1). Furthermore, the hydrothermal pretreatment at lower temperatures increased the proportion of lignin in the residue due to a reduction in the amount of hemicelluloses. It was in agreement with previous study which showed that lignin content in the solid residue is highly affected by the process temperature and could be increased by solubilization of hemicelluloses in the liquid phase (Hansen et al., 2004). However, no major change in Klason lignin was found after hydrothermal pretreatment with or without addition of H_2SO_4 at higher temperatures. Similar results were reported by Monlau et al. (2012) for thermal pretreatment of sunflower stalks at 170 °C for 1 h.

On the contrary to hydrothermal pretreatment, the organosolv pretreatment of stalks at different temperatures and reaction times resulted in the partial removal of lignin. The isopropanol organosolv pretreatment at 160 °C for 30 min with 1% H_2SO_4 caused the highest lignin removal (20.9%), achieving a pretreated substrate with 21.2% Klason lignin (Table 2). Moreover, prolongation of the process from 30 to 60 min at 160 °C had no effect on lignin removal. Contrarily, the pretreatment at 180 °C for 30 and 60 min with 1% H_2SO_4 resulted in 9.3% and 7.1% lignin removal, respectively (Table 2). In other words, increasing the pretreatment temperature from 160

Table 2
Chemical composition and solid recovery of organosolv pretreated sunflower stalks.

Pretreatment conditions				Composition			
T (°C)	Time (min)	H_2SO_4 (%)	Glucan (%)	Xylan (%)	Arabinan (%)	Klason lignin (%)	Solid recovery (%)
140	30	–	48.4 ± 0.5	20.6 ± 0.5	0.9 ± 0.3	26.7 ± 0.4	68.4 ± 0.8
140	60	–	49.1 ± 0.7	19.2 ± 0.8	0.8 ± 0.3	26.4 ± 0.4	64.8 ± 0.2
140	30	1	53.4 ± 0.4	18.9 ± 0.4	0.6 ± 0.2	23.9 ± 0.4	63.2 ± 0.3
140	60	1	54.2 ± 0.6	18.1 ± 0.4	0.3 ± 0.1	23 ± 0.4	60.1 ± 0.1
160	30	–	51.8 ± 0.4	18.3 ± 0.6	0.5 ± 0.1	25.6 ± 0.3	62.1 ± 0.6
160	60	–	52.9 ± 0.3	17.8 ± 0.3	0.4 ± 0.1	25.1 ± 0.4	60.3 ± 0.5
160	30	1	59.6 ± 0.5	17.6 ± 0.3	–	21.2 ± 0.3	56.2 ± 0.5
160	60	1	61.3 ± 0.5	15.8 ± 0.5	–	21.2 ± 0.5	53.4 ± 0.3
180	30	–	56.9 ± 0.4	17.4 ± 0.4	–	22.3 ± 0.5	57.2 ± 0.4
180	60	–	58.9 ± 0.6	17.1 ± 0.4	–	21.9 ± 0.3	55 ± 0.5
180	30	1	66.6 ± 0.6	6.6 ± 0.5	–	24.3 ± 0.6	49.1 ± 0.7
180	60	1	67.8 ± 0.9	5.8 ± 0.2	–	24.9 ± 0.6	46.6 ± 0.6
200	30	–	64.1 ± 0.4	10.0 ± 0.3	–	22.8 ± 0.7	
200	60	–	65.3 ± 0.6	9.1 ± 0.2	–	22.6 ± 0.8	
200	30	1	69.1 ± 0.4	4.9 ± 0.4	–	23.9 ± 0.4	
200	60	1	70.2 ± 0.4	4.0 ± 0.4	–	23.6 ± 0.6	

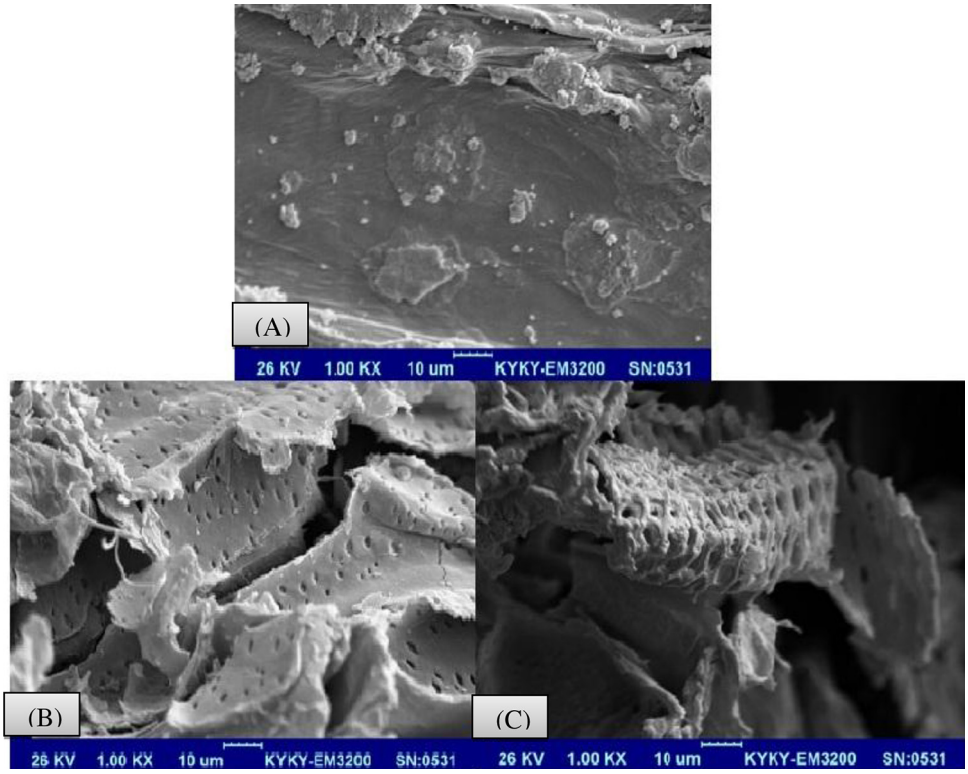

Fig. 1. SEM images of (A) untreated sunflower stalks, (B) hydrothermal pretreated sunflower stalks at 180 °C for 60 min with 1% w/w (based on dry stalks) H_2SO_4, (C) organosolv pretreated sunflower stalks at 160 °C for 30 min with 1% w/w (based on dry stalks) H_2SO_4.

to 180 °C and residence time from 30 to 60 min resulted in lower lignin removal. Therefore, milder conditions were more favorable for lignin removal. It was in line with previous studies, in which the lignin removal is highly influenced by the process temperature and residence time (Amiri et al., 2014; Shatalov and Pereira, 2005). Moreover, the pretreatment at 180 °C for 60 min without H_2SO_4 resulted in 18.3% lignin removal (Table 2). Therefore, addition of 1% H_2SO_4 revealed a positive impact on the lignin removal through the current process. On the other hand, ethanol-based organosolve pretreatment resulted in a higher lignin removal with respect to the isopropanol-based pretreatment (Hideno et al., 2013; Park et al., 2010; Sannigrahi et al., 2010). It has been suggested that the delignification of the lignocellulosic materials is carried out by splitting of lignin–lignin and lignin-carbohydrate bonds and the solubilization of lignin by the organic solvents (Hideno et al., 2013).

In general, the organosolv pretreatment of sunflower stalks resulted in the enhancement of carbohydrate content due to the partial solubilization of lignin. Pretreatment at 160 °C for 30 min with 1% H_2SO_4 resulted in the highest increase in carbohydrate content (28%), while the pretreatment without H_2SO_4 at the severe conditions, i.e., 180 °C for 60 min, resulted in the material with 26% higher carbohydrate content compared with untreated stalks (Table 2). Furthermore, xylan and arabinan, derived from hemicelluloses, were decreased or totally vanished in all organosolv pretreated samples with 1% H_2SO_4 (Table 2). These results show that sulfuric acid is an active catalyst for the degradation of xylan and arabinan from hemicelluloses in sunflower stalks.

The morphological features of the pretreated and untreated stalks were studied by SEM images to obtain perceptions into the surface characteristics. Significant morphological changes can be observed in the pretreated substrates compared to the untreated biomass (Fig. 1). As can be seen in Fig. 1, the untreated sunflower stalks had a compact solid structure, while the pretreated samples had a sparse and rough shape with more fragile bundles. The pretreated samples appeared to be more fragile and exhibited more sponge-shape with more internal surfaces compared to the untreated samples, which can facilitate further bacterial accessibility (Bateni et al., 2014). Therefore, the effects of pretreatments on the consecutive biodegradation of the cellulose rich fractions need further investigations.

The structural changes in sunflower stalks were analyzed by FTIR spectroscopy. The spectra and the absorption data of the untreated sunflower stalks and the pretreated samples with the highest methane production are presented in Fig. 2. The absorbance at 1430 and 898 cm^{-1}, representing crystalline cellulose I and cellulose II, respectively, were used to study crystallinity changes (Colom et al., 2003). The absorbance ratio of A_{1430}/A_{898}, defined as crystallinity index (CI), was decreased from 0.78 for the untreated substrate to 0.75 and 0.67 for the pretreated samples by the hydrothermal (at 180 °C for 60 min with 1% w/w H_2SO_4) and organosolv at 160 °C for 30 min with 1% w/w H_2SO_4) pretreatments, respectively.. In other word, cellulose crystallinity was decreased by 14.1% and 3.8% by organosolv and hydrothermal pretreatments, respectively.

3.3. Biogas production

The untreated and pretreated sunflower stalks were subjected to a batch anaerobic digestion for biogas production by a mixed culture for 45 days. Fig. 3 represents the accumulated methane yield of sunflower stalks pretreated at different temperatures and retention times (without H_2SO_4) as well as the untreated stalks. As can be seen, the methane yield of 124 mL/g VS was obtained from untreated sunflower stalks after 45 days anaerobic digestion, which is about 35% and 48% lower than the reported yield by Monlau et al. (2012) and Antonopoulou et al. (2010), respectively. This may be attributed to differences in the chemical composition of sunflower stalks or microbial compositions of the digesting sludge. Both

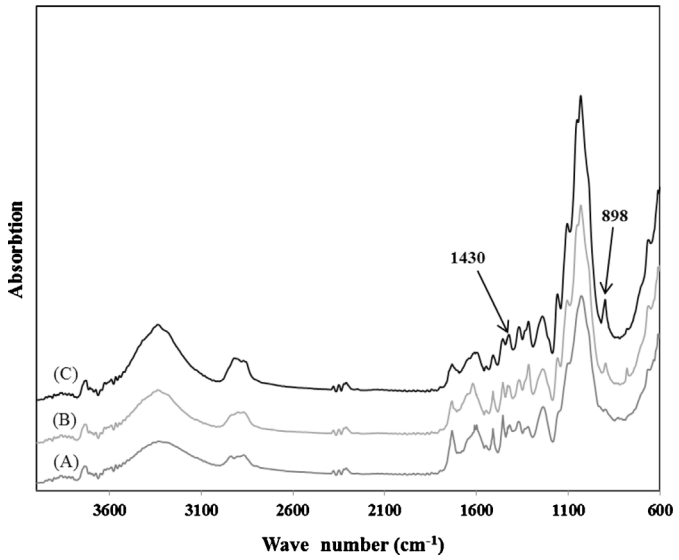

Fig. 2. FTIR spectra of (A) untreated sunflower stalks, (B) hydrothermal pretreated sunflower stalks at 180 °C for 60 min with 1% w/w (based on dry stalks) H_2SO_4, (C) organosolv pretreated sunflower stalks at 160 °C for 30 min with 1% w/w (based on dry stalks) H_2SO_4.

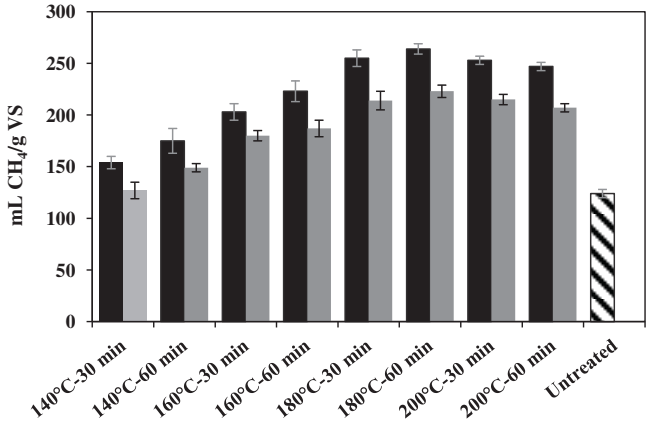

Fig. 3. Yield of accumulated methane production (mL/g VS) after 45 days of anaerobic digestion of untreated (shaded bar), hydrothermal pretreated (grey bars), and organosolv pretreated (black bars) sunflower stalks (error bars are standard deviation).

the hydrothermal and the organosolv pretreatments increased the biomethane yield of the sunflower stalk. Increasing the temperature of both hydrothermal and organosolv pretreatments from 140 to 180 °C had positive impact on the biogas yield. Furthermore, prolongation of the hydrothermal pretreatment from 30 to 60 min led to a slightly higher biogas production yield. The biomethane production yield was significantly affected by the pretreatment temperature; however, prolongation of the pretreatment did not show a significant effect on the biomethane production. In the case of the organosolv pretreatment, the effect of pretreatment time was more significant for the treatment up to 180 °C. The effect of the pretreatment temperature on the biogas production yield was significant but the effect of pretreatment time was not significant. However, for the same analysis on the first three temperatures (140, 160, and 180 °C), both pretreatment temperature and time revealed the significant effects on the biomethane production. These results are in agreement with previous studies showed that the production yield of biomethane and other biofuels such as ethanol from lignocellulosic materials is directly affected by the hydrothermal and organosolv pretreatment time and temperature

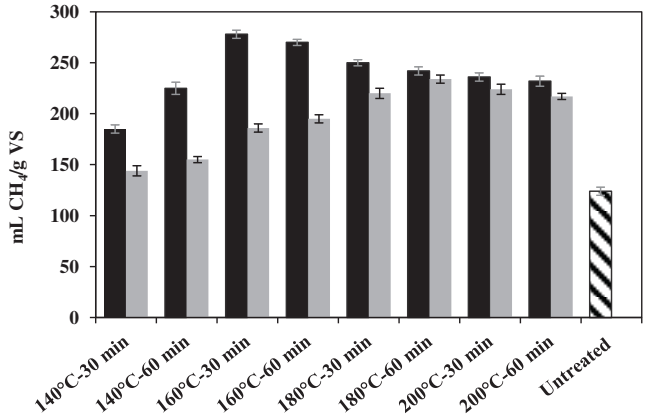

Fig. 4. Yield of methane production (mL/ g VS) after 45 days of anaerobic digestion of untreated (shaded bar), hydrothermal pretreated with 1% w/w (based on dry stalks) H_2SO_4 (grey bars), and organosolv pretreated with 1% w/w (based on dry stalks) H_2SO_4 (black bars) sunflower stalks (error bars are standard deviation).

(Amiri et al., 2014; Chandra et al., 2012; Fernández-Cegrí et al., 2012; Koo et al., 2011). The highest methane production yield of 223 and 264 mL/g VS were achieved by the hydrothermal and organosolv pretreated sunflower stalks at 180 °C for 60 min, respectively. Therefore, organosolv pretreatment was more effective than hydrothermal pretreatment for biomethane production from sunflower stalks which may be due to a higher total carbohydrate content with less lignin content in the organosolv pretreated stalks. Increasing the temperature of both hydrothermal and organosolv pretreatments from 180 to 200 °C slightly decreased the methane production yield. This may be due to the enhanced solubilization of hemicelluloses (xylan and arabinan) in liquid fraction.

Fig. 4 depicts the effect of hydrothermal and organosolv pretreatments in the presence of H_2SO_4 on the methane production yield from the sunflower stalks. Addition of 1% w/w (based on dry stalks) H_2SO_4 not only showed positive effect in the final methane production yield but also resulted in the highest methane production yields to be occurred in milder conditions. The analysis of variance showed the significant effect of the presence of acid on the results. The best results of biomethane production were obtained from the organosolv pretreated stalks at 160 °C for 30 min in the presence of H_2SO_4, where 278 mL methane/g VS was obtained after 45 days (Fig. 4). This implied 124% improvement in the methane yield compared to the untreated stalks. Furthermore, the maximum methane production yield from hydrothermal treated stalks in the presence of H_2SO_4 was obtained at 180 °C for 60 min equal to 234 mL/g VS (Fig. 4). These results indicated that organosolv pretreatment was more influenced by addition of H_2SO_4 in the improvement of methane yield compare to the hydrothermal pretreatment. The results also showed that increasing the organosolv pretreatment temperature (in the presence of H_2SO_4) from 160 to 200 °C slightly decreased the methane production yield. This may be due to remarkable hemicellulose removal at the elevated temperature in the presence of the acid, even though the glucan content increased. These results indicated that methane production yield was more dependent on hemicellulose compared to cellulose. The methane and carbon dioxide distribution in the produced biogas were reported in Table 3. Carbon dioxide percentage was within a range of 32-52%. The pretreated substrate at higher temperature (up to 180 °C) resulted in lower carbon dioxide content in the biogas. Although the presence of the sulfuric acid led to lower carbon dioxide content in the digested organosolv pretreated stalk, it did not show any trend for the hydrothermal pretreated samples.

The maximum biomethane production yield of 278 mL/g VS obtained in this work was higher than the yields previously

Table 3
Methane and carbon dioxide distribution in the produced biogas samples.

Pretreatment conditions			Hydrothermal pretreatment		Organosolv pretreatment	
T (°C)	Time (min)	H_2SO_4 (%)	CH_4 (%)	CO_2 (%)	CH_4 (%)	CO_2 (%)
140	30	–	53	47	49	51
140	60	–	51	49	48	52
140	30	1	53	47	56	44
140	60	1	55	45	57	43
160	30	–	56	44	52	48
160	60	–	61	39	53	47
160	30	1	55	45	68	32
160	60	1	62	38	67	33
180	30	–	64	36	60	40
180	60	–	65	35	61	39
180	30	1	64	36	64	36
180	60	1	65	35	61	39
200	30	–	62	38	61	39
200	60	–	62	38	62	38
200	30	1	63	37	61	39
200	60	1	61	39	60	40

reported through different pretreatments of sunflower stalks. Anaerobic digestion of sunflower stalks by mixed culture using three thermal pretreatments at 55 °C for 24 h, 170 °C for 1 h, and 121 °C for 60 min resulted in the production of 198, 219, and 270 mL/g VS methane, respectively (Antonopoulou et al., 2010; Monlau et al., 2012). Thermo-chemical pretreatment of sunflower stalks using NaOH (4%; 55 °C; 24 h), H_2O_2 (4%; 55 °C; 24 h), $Ca(OH)_2$ (4%; 55 °C; 24 h), $FeCl_3$ (10%; 170 °C; 1 h), and HCl (4%; 170 °C; 1 h) led to the methane yield of 259, 256, 241, 248, and 233 mL/g VS, respectively (Monlau et al., 2012). In addition, anaerobic digestion of sunflower stalks pretreated in commercial-scale by hot-water maceration (75–95 °C for 20-200 sec) followed by steam-explosion (0.8-2.2 MPa; 0.3 L; 2–20 min) resulted in the methane yield of 72.3 mL/g VS (Maroušek, 2013).

4. Conclusions

Both hydrothermal and organosolv pretreatment of the sunflower stalk enhanced methane production yield. However, the organosolv pretreatment led to more promising results at the same operating conditions (temperature and duration). The presence of 1% sulfuric acid significantly enhanced the effectiveness of the hydrothermal pretreatment. The highest methane yield of 234 mL/g VS was achieved from the hydrothermal pretreated stalk at 180 °C for 60 min. On the other hand, organosolv pretreatment in the presence of sulfuric acid is more favorable at milder conditions. The most promising result for methane production was as high as 278 mL/g VS for the organosolv pretreated at 160 °C for 30 min in the presence of 1% sulfuric acid.

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