Enhanced solid-state anaerobic digestion of lignocellulosic biomass by organosolv pretreatment

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Abstract

Lignocellulosic materials are abundant and renewable feedstocks of bioenergy which has recently been used for production of the so-called second-generation biofuels. Pretreatment process is an essential stage to improve the digestibility of lignocellulosic substrates. In this paper, an organosolv process was used to improve the methane yield by solid-state anaerobic digestion (SSAD) of three lignocellulosic substrates (elm, pine wood, and rice straw). To our knowledge, there is no publication on using organosolv pretreatment prior to SSAD. The unique advantage of the organosolv pretreatment is to separate lignin as a value added by product. The Organosolv pretreatment was conducted in four different conditions (at 150 and 180 °C for 30 and 60 min) using 75% ethanol solution on the lignocellulosic materials and the methane production yield through the SSAD was investigated. The results showed that the total methane yield of the pretreated elm, pine, and rice straw was enhanced by 90, 83, and 36%, respectively. The effects of the pretreatment temperature and time on methane yield were also investigated. Statistical analyses showed that the pretreatment temperature was the most influencing factor in the SSAD, while the effect of pretreatment time on methane production from elm, pine, and rice straw was not significant. Almost all of the substrates produced biogases with methane contents between 40% and 50% between day 14 and day 55.

Keywords: Biogas, Solid-state anaerobic digestion, Organosolv pretreatment, Rice straw, elm wood, Pine wood.

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1. Introduction

Worldwide concerns about limitation of fossil resources, rising crude oil prices, and greenhouse gas (GHG) emissions lead researchers to find alternative clean and renewable energy sources [1]. Lignocellulosic materials are abundant and renewable feedstocks of bioenergy which has recently been used for production of the so-called second-generation biofuels [2-5]. Compared to the most of liquid biofuels, biogas, as a second-generation biofuel, has been shown to have a far better performance with regard to both agricultural land area efficiency and life cycle emissions [6]. Biogas produced during an anaerobic digestion (AD) of lignocellulosic materials [7], can be used as a versatile source of energy to produce heat, electricity, and combined heat and electricity. Moreover, it offers other advantages like controlling organic wastes, reducing greenhouse gas emission, and producing a proper fertilizer [8, 9]. AD processes are classified based on the solid content of media into liquid anaerobic digestion (L-AD), and solid-state anaerobic digestion (SSAD) [10]. L-AD operates at a total solid (TS) content of less than 15% while SSAD is generally used at a TS content of higher than 15% [11]. Smaller specific reactor volume, fewer moving parts, lower energy input for heating, easier to handle the end-product, and lower parasitic energy loss are the main advantages of SSAD in comparison with L-AD [11-13]. SSAD is specially matched well with lignocellulosic feedstocks such as agricultural that are available in low moisture content forms [11, 14]. However, the anaerobic digestion of lignocellulosic substrates, specially SSAD, is limited by the rate of hydrolysis due to the substrate recalcitrant structure [15]. Therefore, pretreatment process is an essential stage to improve the digestibility of lignocellulosic substrates through SSAD [15, 16].

Although different affecting factors, e.g., the crystallinity of cellulose and accessible surface area, may play important roles in the bioconversion of lignocelluloses, the presence of lignin is apparently the most important factor affecting the biodegradability of lignocelluloses [17-20]. The lignin-carbohydrate matrix limit the digestibility of lignocelluloses since lignin is a cross-linked network hydrophobic polymer that remains insoluble in all solvents and is fairly resistant to enzymatic and microbial degradation [21, 22]. Therefore, both yield and rate of biomethane production from lignocelluloses can be improved by delignification of lignocellulosic materials. Ethanol organosolv process is one of the most promising pretreatments which improve the bioconversion of lignocelluloses by extraction of lignin [23, 24]. Furthermore, the unique advantage of the organosolv pretreatment is to separate lignin as a value added by product [24]. Therefore, using ethanol organosolv pretreatment prior to the AD process could improve the economy of the process both by increasing methane yield in SSAD systems and separating lignin as a value added product [25]. To our knowledge, there is no publication on using organosolv pretreatment prior to SSAD.

In this study, ethanolic organosolv pretreatment was evaluated to improve the solid-state fermentation of three lignocellulosic materials, i.e., elm, pine, and rice straw. The main objective of this research was to determine effects of the organosolv pretreatment on the methane yields and biogas compositions during SSAD. Effects of the main pretreatment parameters, i.e., temperature and time, on the methane yield were determined.

2. Material and methods

2.1. Feedstocks and inoculum

Elm, as a hardwood, pine as a softwood, and rice straw as an agricultural waste, were used for biogas production. Elm and pine, obtained from Isfahan University of Technology Forest (Isfahan, Iran), and rice straw was prepared from Lenjan field with a cultivar named "Sazandegi" (Isfahan, Iran). Elm and pine woods were debarked, cut into smaller pieces, and milled to obtain chips with size of less than 2 cm. The chips and straw were partly ball-milled and screened to achieve powder with a size between 295 and 833 μm (20–80 mesh). The screened substrates were then stored at room temperature in resealable plastic bags until use. Effluent from a 7000 m^3 mesophilic anaerobic digester (Isfahan municipal sewage treatment, Isfahan, Iran) was used as an inoculum. Due to the low total solid (TS) content, the inoculum was centrifuged at 4500 rpm for 30 min to obtain desirable TS content. The supernatant was discharged, and the remaining sludge was mixed to obtain a homogenous inoculum for SSAD. Inoculum was starved for 1 week to remove the degradable volatile solids (VS). Cellulose, hemicellulose, lignin, TS, and VS contents of the feedstocks and inoculum were analyzed.

2.2. Organosolv pretreatment

Ethanol as an organic solvent and sulfuric acid as a catalyst were used for the pretreatment. Each feedstock was mixed with 75% (v/v) aqueous ethanol solution supplemented with 1% w/w (based on dry mass) sulfuric acid with solid-to-liquid ratio of 1:8 (based on dry mass). Treatments were carried out in a 500 mL high-pressure stainless steel batch reactor [26]. The reactor was heated at a rate of 3 °C/min to the desired temperature, i.e., 150 or 180 °C, and the temperature was controlled to be constant for 30 or 60 min. Afterwards, the reactor was cooled in an ice bath. The pretreated substrates were then washed three times with 100 mL aqueous ethanol (75% v/v, 60 °C) and air dried overnight. Finally, the pretreated materials were stored in resealable plastic bags at room temperature until use.

2.3. Solid-state anaerobic digestion (SSAD)

The untreated and pretreated samples of the elm, pine, and rice straw were mixed with an appropriate amount of inoculum and deionized water to achieve feed to inoculum ratio (F/I) (based on VS) of 3 at the initial TS content of 21%. The mixed materials, in the 118 mL glass reactors, were incubated in a convection oven (JSH20LURS, JAHL Co., Karaj, Iran) for up to 55 days at mesophilic conditions (39 ± 1 °C). Inoculum without any substrate was evaluated as a control. Anaerobic conditions were provided by purging the reactors with nitrogen gas for about 2 min. All the digesting experiments were run in duplicate. Gas samples were taken every 3 days during the first 9 days and every 5 or 6 days until 55 days and analyzed for biogas composition and volume.

2.4. Analytical methods

TS and VS contents of the feedstocks and inoculum were measured through the drying at 105 °C followed by heating at 575 °C to a constant weight [17]. The untreated and pretreated samples were analyzed for carbohydrates and lignin contents according to the method presented by Sluiter et al. [27]. Concentration of the sugars was analyzed by HPLC (Jasco International Co., Tokyo, Japan), equipped with a RI detector and an ion-exchange Aminex

HPX-87P column (Bio-Rad, Richmond, CA, USA) at 80 °C with 0.6 ml/min flow rate of deionized water as a mobile phase.

Biogas volume and composition were determined every 3 to 6 days for each digester. Methane and carbon dioxide produced in the anaerobic digestion were analyzed using a gas chromatograph (Sp-3420A, TCD detector, Beijing BeifenRuili Analytical Instrument Co., China) equipped with a packed column (3 m length and 3 mm internal diameter, stainless steel, Porapak Q column, Chrompack, Germany). The carrier gas was nitrogen operated with 45 mL/min flow rate. Temperature of the column, injector, and detector were 40, 100, and 150 °C, respectively. A pressure-tight syringe (VICI, Precision Sampling Inc., USA) with volume of 250 µL was used for gas sampling and injection, making it possible to take the gas samples at the actual pressure of the bioreactors. The excess gas was released through a needle after each gas sampling to avoid the overpressure in the bottles. All the experiments were performed in duplicates, and the averages of the results are presented.

2.5. Statistical analysis

Statistical significance was determined by analysis of variance (ANOVA) using Minitab software (Version 15) with a threshold p-value of 0.05.

3. Results and discussion

3.1. Characterization of inoculum

The inoculum from the mesophilic AD was centrifuged to increase its solid content. As shown in Table 1, the TS and VS contents were increased from 5.69 and 2.75 to 11.75% and 5.35%, respectively, as a result of the centrifugation. Centrifugation was also used to maintain the TS contents of all reactors around 20% [28].

Table 1. Characteristics of feedstocks and inoculum.

Samples	Pretreatment	TS content (%)	VS content (%)	Lignin (%)	Cellulose (%)	Hemicellulose (%)
Inoculum	Untreated	5.69	2.75	ND	ND	ND
	Centrifuged	11.75	5.35	ND	ND	ND
Elm	Untreated	95.5	94.5	26.54	48.40	25.06
	150°C, 0.5h	95.5	94.1	25.13	45.28	29.59
	150°C, 1h	95.5	93.8	23.19	44.71	32.10
	180°C, 0.5h	96.3	94.4	21.17	54.10	24.73
	180°C, 1h	94.9	93.6	20.50	50.70	28.80
Pine	Untreated	95.1	95.2	26.55	45.08	28.37
	150°C, 0.5h	95.3	94.6	27.15	47.57	25.28
	150°C, 1h	95.9	95.1	26.63	53.16	20.21
	180°C, 0.5h	96.5	95.5	21.73	68.83	9.44
	180°C, 1h	96.9	95.8	21.10	68.06	10.84
Rice straw	Untreated	95.4	83.9	17.09	49.17	33.74
	150°C, 0.5h	95.6	83.8	14.03	55.62	30.35
	150°C, 1h	95.7	83.6	13.40	54.76	31.84
	180°C, 0.5h	95.9	86.2	11.46	58.87	29.67
	180°C, 1h	96.0	84.7	11.14	62.29	29.57

ND = not determined.

3.2. Pretreatment

Elm, pine, and rice straw were subjected to ethanol organosolv pretreatment prior to anaerobic digestion in order to improve the yield of biomethane production. The pretreatment was conducted at 150 and 180 °C for 30 and 60 min using 75% aqueous ethanol containing 1% w/w (based on raw material dried mass) sulfuric acid as a catalyst. The solid content in the form of TS, VS, as well as lignin, cellulose, and hemicellulose contents of the untreated and pretreated materials were determined and summarized in Table 1.

Considering the untreated materials, lignin content of pine and elm was 55% higher than rice straw. The untreated elm, pine, and rice straw consisted of 48.4, 45.1, and 49.2% glucan, respectively. Different parts of materials were differently affected by the pretreatment. Depending on the pretreatment conditions, 5 to 23% of lignin content of elm, 0.3 to 21% of that of pine, and 18 to 35% of that of rice straw were removed through the pretreatment. Increasing the severity of pretreatment generally resulted in higher lignin removal. Considering the composition of the materials, pretreatment of pine was affected by the temperature more than elm and rice straw. A relatively high portion of lignin (21%) and hemicellulose (62%) of pine was removed through the pretreatment at 180 °C for 60 min. As a result, a pretreated pine wood with more than 68% cellulose content was achieved through the pretreatment. Organosolv pretreatment of elm and rice straw, at 180 °C for 60 min, resulted in 23% and 35% lignin removal, respectively, through which the pretreated material with 51% and 62% cellulose was obtained.

3.3. Biogas production

Organosolv pretreatments in four different conditions were performed on the lignocellulosic materials, and the methane production yield through the SSAD was investigated. The total methane production yield from untreated and pretreated materials is shown in Fig.1.

Considering the methane production from the untreated materials, the highest methane yield of 99.2 $L.kg^{-1}$ VS was obtained from rice straw that had the lowest lignin content. The digestion of untreated elm and pine resulted in production of methane with the yields of 41.8 and 29.5 $L.kg^{-1}$ VS, respectively. The presence of pores in the structure of hardwoods which facilitate the accessibility of microorganisms might be responsible for the higher yield obtained from elm in comparison to pine [29].

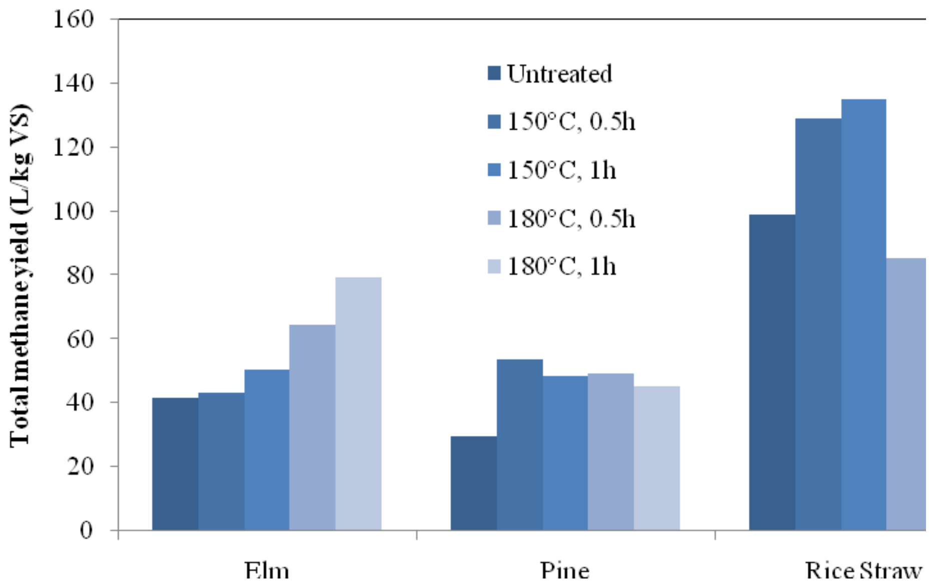

Fig. 1. Effect of pretreatment temprature and time on total methane yield (digestion time: 55 days, TS: 21%, F/I = 3).

The yields of methane production from the woods were generally improved by the pretreatment at all conditions, whereas the yield from rice straw was only improved by the pretreatment at the lower temperature of 150 °C. The highest methane yield of 135.2 $L.kg^{-1}$·VS was obtained from the rice straw pretreated at 150 °C for 1 h. Even though methane production from elm was improved by increasing the pretreatment severity, the yield of methane production from pine was decreased by the pretreatment at high severities. In the case of rice straw, however, the yield of methane production was reduced by increasing the pretreatment temperature but increased with the time of the pretreatment. Therefore, pretreatment of rice straw at the higher temperature affected the methane production in two different ways. Even though the yield of methane production from rice straw was decreased as a result of the pretreatment at high temperature, prolonging the pretreatment improved the methane production. The highest yield of methane production from pine and rice straw were 53.9 and 135.1%, respectively, which were obtained through the pretreatment of pine at 150 °C for 0.5h and rice straw at 150 °C for 1h. . The maximum increase in the yield of methane production, 90.45 %, was obtained after the organosolv pretreatment of elm at 180 °C for 1 h. The pretreatment of pine at 150 °C for 0.5 h and rice straw at 150 °C for 1 h resulted in the improvement of the yield up to 82.8 and 36.2 %, respectively. Even though increasing the pretreatment temperature from 150 °C to 180 °C resulted in improvement of the yield of methane production from elm, it showed negative effects on methane production from pine and rice straw. Although the pretreatment of pine had a remarkable effect on the yield of methane production (54 to 83% improvement), the pretreatment conditions, i.e., temperature and time, with p-values of 0.28 and 0.91, respectively, did not showed significance effects on the methane yield. Overall, the pretreatment temperature had a significant effect on the methane production from elm and rice straw, while the effect of pretreatment time on methane production from elm, pine, and rice straw was not significant (p- value of 0.14, 0.91, and 0.27, respectively).

Daily methane yields during SSAD of the lignocellulosic materials are shown in Fig. 2. The daily methane production was highly dependent on the pretreatment conditions, while it generally showed two peaks at the about days 6th and 14th. The daily methane production at the peak points was improved by the organosolv pretreatment at the proper conditions. Pretreatment of elm at 180 °C for 1 h, pine at 180 °C for 0.5 h, and rice straw at 150 °C for 0.5 h resulted in 224, 153, and 25 % increase in the daily methane production at day 6th. At the second peak, day 14th, however, the pretreatments resulted in 65, 91, and 51 % increase in the methane production from elm, pine, and rice straw, respectively. Methane production from pine and elm was significantly diminished after the peaks. In the case of rice straw, however, appreciable amounts of methane were produced after the peaks, from day 20th to 40th. In addition, the pretreatment of rice straw negatively affect the methane production after the peaks. However, methane production through the anaerobic digestion of rice straw pretreated at 150 °C for 1 h was more stable than that of the other pretreated materials, in the first 15 days.

Methane contents in the accumulated biogas produced by SSAD from elm wood, pinewood, and rice straw are shown in Fig. 3. Almost all of the substrates produced biogases with methane contents between 40% and 50% between day 14 and day 55 with the exception of the pretreated elm at 150 °C for 1 h which had the highest final methane content (63 %).

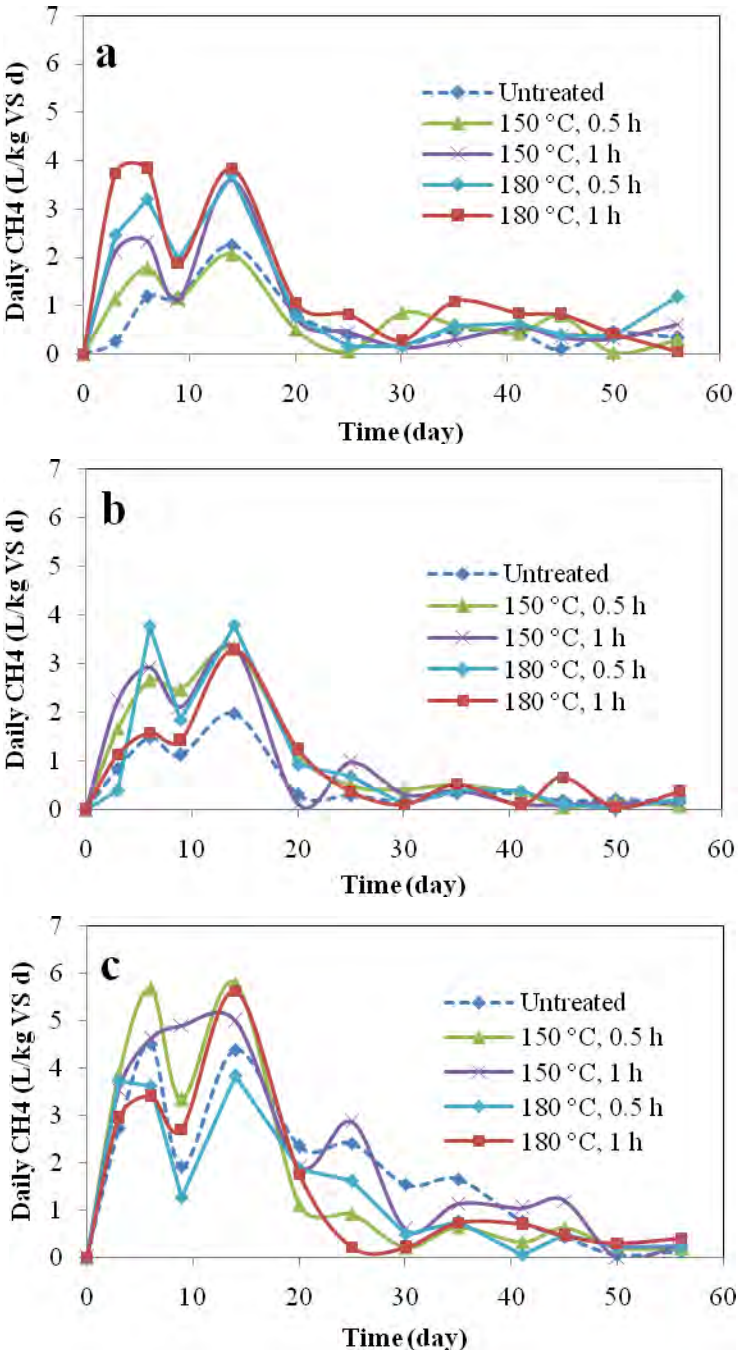

Fig. 2. Total methane production during 55-day SSAD of a) elm wood, b)pine wood, and c)rice straw.

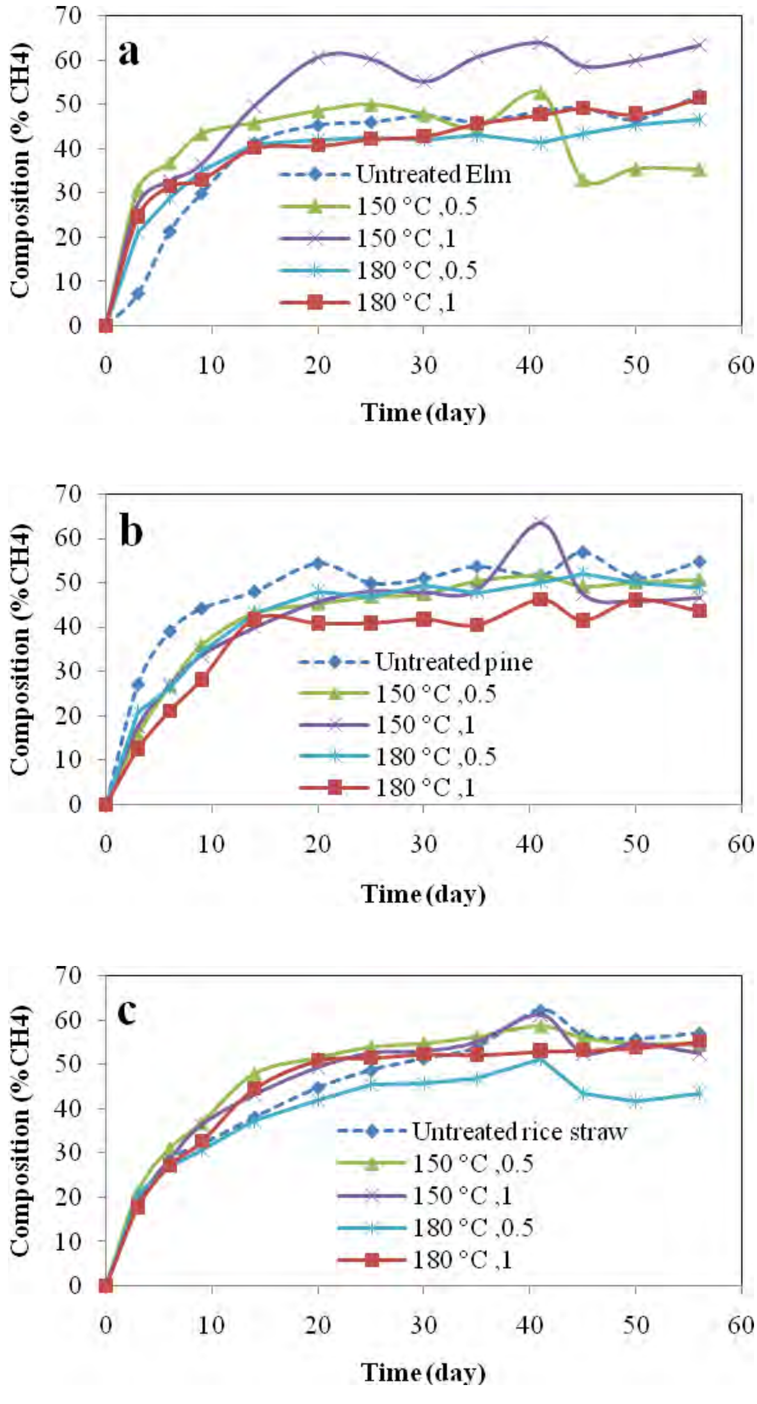

Fig. 3. Methane contents in biogas produced during SSAD of untreated and pretreated a) elm wood, b) pine wood, and c) rice straw in different pretreatment conditions.

4. Conclusion

Organosolv pretreatment can be used for improvement of methane production from lignocellulosic materials by SSAD. The organosolv pretreatment of elm, pine, and rice straw using proper pretreatment conditions resulted in 90, 83, and 36% increase in the total methane yield by SSAD, respectively. The highest methane yields of 135.2 $L.kg^{-1}\cdot VS$ rice straw, 79.5 $L.kg^{-1}\cdot VS$ elm, and 53.9 $L.kg^{-1}\cdot VS$ pine were obtained using the organosolv pretreatment.

The highest final methane content was 63 %, observed from the pretreated elm at 150 °C for 1 h.

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