Effect of particle size and hydration treatment on the wheat straw biodegradability and hydrogen production by a microbial consortium.

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ABSTRACT

Lignocellulosic materials are promising for the production of biofuels since they are abundant and widely distributed. However, their biodegradability is low since they are composed of polymers of cellulose and hemicellulose tightly bound to lignin. In order to face up to this challenge this work study the effect of hydration treatment and particle size on the biodegradability of raw wheat straw measured as hydrogen production by a microbial consortium. Dry wheat straw was milled at 3mm and 0.2mm and the hydration treatments were performed until reach the maximum water retention capacity with water and effluents from a H_2-producing reactor. Then, the hydrated wheat straw was loaded into 100 ml reactors at 25g ST/L using 10% sludge from an anaerobic digester as inoculum and incubated at 37°C. According to the results obtained, the hydration treatment (p<0.01) and particle size (p<0.5) had a significant effect on the biodegradability of wheat straw measured as hydrogen production (ml H_2/L) and hydrogen production rate (ml H_2/L·d). For the particle size, the hydrogen production was higher at 3mm than that at 0.2mm mainly for untreated wheat straw. On the other hand, hydration treatments with water or effluents displayed similar hydrogen production rates of 39 ml H_2/L·d being 60% higher than those for untreated wheat straw. The highest hydrogen production was reached with the effluent-hydrated wheat straw in comparison with the water-hydrated wheat straw and control (366, 293 and 193 ml H_2/L, respectively). The hydration treatment with water or effluent is an economical method to increase effectively the biodegradability of lignocellulosic materials when anaerobic mixed consortia are used.

Keywords: hydration, water retention capacity, wheat straw.

1. Introduction

The decrease of oil reserves, the high costs of fuels and the global interest for the reduction of effects of GHG have encouraged the demand for alternative energy that can help to the gradual shift of fossil fuels. The hydrogen is an energy carrier that could help to substitute the requirements of global energy due to its high combustion power (H_2 >120MJ/Kg vs. fossil fuels > 45 MJ/kg). Hydrogen is considered as a green fuel since the water is the only product at the end of its theoretical combustion [1]. It can be produced by biological processes; a common one includes the conversion of carbohydrates into hydrogen by the action of fermentative microorganisms in anaerobic environments. One of the main advantages of using microorganisms for hydrogen production is the reutilization of wastes generated by the industrial activity, where the biomass constitutes a key component of them. Furthermore, GHG are reduced due to raw material has previously fixed carbon for its growth [2]. For this purpose, natural environment isolated or genetic modified microorganisms can be utilized, both can growth in refined carbohydrates such as glucose and sucrose. Another type of microorganisms includes mixed cultures or microbial consortia, which can be used due to their ability to growth in complex substrates such as wastewater, solid wastes, anaerobic sludge and agricultural wastes. Particularly, agricultural wastes have a high potential to be utilized for hydrogen production since they are abundant, cheap and a good source of carbohydrates [3]. It's estimated that every year 200 billion of tons of agricultural wastes are produced worldwide being the equivalent to 60-80 billion of tons of crude oil [4]. In Mexico, 150 million of dry matter is produced coming from corn straw, sorghum straw, tops/leaves of sugarcane and wheat straw. Particularly wheat straw is a residue not very exploited and concentrated in some regions of Mexico, accordingly can be collected without difficulty and at low cost for its transformation into bioenergy [5]. The chemical composition of wheat straw shows a high content of fermentable sugars, which are most found as a homopolymer of glucose (cellulose), a heteropolymer of xylose and other sugars of 5 carbons (hemicellulose), strongly bonded by a heteropolymer of aromatic compounds (lignin) [6]. The use of wheat straw for hydrogen production has not yet been extensively studied. Fan et al. (2006) [7] compared the hydrogen production using raw wheat straw and HCl pretreated wheat straw, the maximum cumulative hydrogen production, 68.1 ml H_2/g TVS was observed from HCl pretreated wheat straw which was 136-fold as compared with that of raw wheat straw wastes. Kongjan et al. (2010) [8] produced H_2 from wheat straw hydrolysate by dark fermentation using extreme thermophilic mixed culture, the highest production 318.4 ml H_2/g-sugars, was obtained with 5% of hydrolyzed substrate. Quéméneur et al. (2012) [9] studied the hydrogen production using an anaerobic heat-treated sludge fed with raw and enzymatically hydrolyzed wheat straw. They reported a maximal production of 10.52 and 19.63 ml H_2/g SV, respectively. The optimal dosis of enzyme mixture added to the wheat straw was between 1 – 5 mg/g wheat straw. Panagiotopoulos et al. (2013) [10] shows the hydrogen production by *Caldicellulusiruptor saccharolyticus* from hydrolysates derived from a dilute-acid treatment and enzymatic hydrolysis of wheat grain and straw. The mixed hydrolysates exhibited good fermentability at 20g/L. Nasirian et al. (2011) [11] also reports hydrogen production from acid-pretreated wheat straw, they found that simultaneous saccharification and fermentation (SSF) is the most effective and economical method to convert the substrate into hydrogen reaching a yield of 1 mol/mol-glucose. Additionally, different reactor settings have been studied result the best production rates using an up-flow anaerobic sludge bed (UASB) reactor compared with continuously-stirred tank reactor (CSTR) or anaerobic filter (AF) reactor [12]. Nonetheless it can be notice from these studies; the biodegradability of wheat straw is restricted by the pretreatment utilized (chemical, enzymatic or physical). The main drawback of utilization of pretreatments is that increases the cost of production in a scale-up process. Besides, the chemical pretreatments can produce inhibitors as furfural and hydroxymethylfurfural having to be removed from the hydrolyzed in order to avoid the inhibition of the fermentation [13]. However, there are other alternative pretreatments that can be utilized thanks to its low cost and low energy demand obtaining a higher biodegradability of lignocellulosic substrates. For example, the hydration of fibers is one the initial steps before its solubilization. Recently Lara and Valdez (2013) [14] studied the hydration properties of wheat straw, they found the intervals of time where is obtained the highest water retention capacity (WRC) and sugars release at the same time. In this way, the objective of this work was to study the effect of hydration of wheat straw fibers and particle size on the hydrogen production by a microbial consortium.

2. Experimental Procedure

2.1 Preparation of substrate.

Wheat straw (*Triticum aestivum L.*) was obtained from agricultural wastes in the locality of Irapuato, Guanajuato, Mexico in 2010. Table 1 shows the wheat straw chemical composition.

Table 1. Chemical composition of wheat straw.

Component	
Carbon (g/Kg)	419
Phosphorus (g/Kg)	0.46
Total Kjeldahl nitrogen (g/Kg)	4.4
Total solid volatiles (% ST)	86
Ashes (%ST)	8.6
Fiber content (% ST)	38.7
Protein (g/Kg)	30.6

One kilogram of sample was milled using a balls miller to obtain a particle size of 3 and 0.2 mm. Then, wheat straw was hydrated in water (10 h) and effluents from a biological reactor producing H_2 (14 h). The quantity of water or effluent added to the wheat straw was 20 ml/g wheat straw. The mixture was well-homogenized and kept at room temperature until get the maximum water retention capacity [14]

2.2 Batch reactor operation.

Batch tests for hydrogen production were performed in 100 ml bottles. 14 ml of sludge from an anaerobic digester were mixed with 56 ml of mineral medium (KH_2PO_4 44.8 g/L, $K_2HPO_4{\cdot}3H_2O$ 6.98 g/L, $MgCl_2{\cdot}6H2$ 0.1 g/L, $CaCl_2$ 0.02 g/L, NH_4Cl 6 g/L, $MnSO_4{\cdot}6H_2O$ 0.015 g/L, $FeSO_4{\cdot}7H_2O$ 0.025 g/L, $CuSO_4{\cdot}5H_2O$ 0.005 g/L, $CoCl_2{\cdot}5H_2O$ 0.000125 g/L). The initial pH for hydrogen production was maintained to 5.5 and the hydrated wheat straw (3 mm or 0.2 mm) was added to a concentration of 25 g ST/L. The medium utilized for wheat straw hydration was added to the reactor and the volume was completed with mineral medium. The reactors were incubated statically during 20 days at 37°C. Untreated wheat straw (3 mm and 0.2 mm) was utilized as a control experiment and all experiments were done by triplicate.

2.3 Analytic methods

Biogas volume was determinate using lubricated syringes and the hydrogen content was analyzed with a gas chromatograph (Perkin Elmer Clarus 580) with a thermal conductivity detector equipped with an Elite GC – GS Molsieve capillary column (30 meter, 0.53 mm ID). The injector and detector temperatures were 50°C and 200°C, respectively, the oven temperature 50°C - 100°C; 7°C/min. Argon was utilized as carrier gas at a flow rate of 6 ml/min.

3. Results and Discussion.

3.1 Effect of hydration treatment on H_2 production.

The Figure 1A,B shows the effect of hydration treatment of wheat straw on the hydrogen production by a microbial consortia. On average, the hydration treatment (in water or effluent) had a significant positive effect on the wheat straw biodegradability measured as hydrogen production and hydrogen production rate ($p<0.01$). The maximum hydrogen production was 366 ml/l obtained by using wheat straw hydrated with the biological reactor effluent. This can be explained due by enzymes presents in the biological effluent. The effluents were obtained from an anaerobic reactor fed with wheat straw for hydrogen production [14]. According to the literature, the anaerobic microorganisms in the medium release hydrolytic enzymes such as glycosidases, endoglucanases and xilanases. These enzymes have enzymatic activity on cellulose and hemicellulose, where the main products obtained are glucose and xylose. In this way, there were a positive synergic effect between the bacterial enzymes and the hydration. These can be verified with the treatments with water-hydration only, when the hydrogen production was lower. On the other hand, the hydrogen production rates were also increased by the hydration treatments being 60% higher in comparison with the control.

3.2 Effect of wheat straw particle size on H_2 production

In order to study the effect of particle size on the hydrogen production, wheat straw at 3mm and 0.2 mm were utilized as substrate. Several studies have been realized with different lignocellulosic substrates where the hydrogen production has been dramatically increased by decreasing the particle size, since the superficial area is increased and as result the fibers can be easily processed by the microbial enzymes. However, according to the results of this study the hydrogen production was lower at 0.2 mm compared with those obtained at 3mm; this was particularly clear in the control treatments where the hydrogen production was 66 ml/L H_2 vs. 193 ml/L H_2 for 0.2 and 3 mm, respectively. This may be attributed to the damage of the fiber matrix and collapse of the pores during grinding process being affected the water retention capacity value obtained [14]. There was no significant difference between the H_2 production rates at particle size of 0.2 and 3mm.

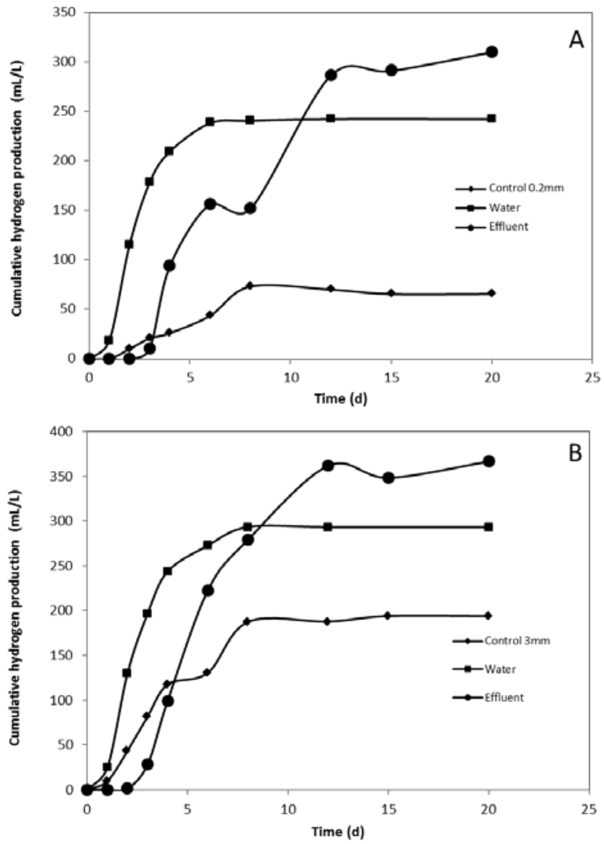

Figure 1. Effect of hydration treatment and particle size on the biodegradability of wheat straw measured as hydrogen production. A) 0.2 mm, B) 3mm.

3.3 Comparison with other studies.

Table 2 shows the comparison of H_2 yields obtained in this work and other studies realized using lignocellulosic feedstocks without chemical or enzymatic pretreatments. In this study, a H_2 yield of 7.4 ml H_2/g ST was obtained from the non-hydrated wheat straw. This result is similar to those reported by Quéméneur et. al. (2012) who used similar conditions such as incubation temperature, inoculum and untreated wheat straw. On the other hand, a H_2 yield of 14.6 ml H_2/g ST was obtained in this study from a hydrated substrate; this yield was similar to those reported for the most thermophilic systems except for Li and Liu (2012) who reached a very high yield by using pure cultures.

Table 2. Hydrogen yield obtained from lignocellulosic feedstocks without pretreatment.

Substrate	Inocula	Incubation temperature	H_2 yield (ml H_2/g ST)	References
Hydrated wheat Straw	Anaerobic digester	37°C	14.6	This study
Untreated wheat straw			7.4	
Wheat straw	Anaerobic sludge	37°C	9.2	Quéméneur et. al 2012
Rice straw	Wastewater sludge	55°C	9	Chen et. al 2012
Rice straw	Sewage sludge	55°C	15.2	Kim et. al 2013
Rice straw	Sewage sludge	55°C	18.0	Kim et.al, 2012
Cornstalk waste	*C. thermocellum* DSM 7072 and *C. thermosaccharolyticum* DSM 869	55°C	68.2 74.9	Li and Liu, 2012
Wheat straw	*Thermophilic strain isolated from hot spring*	65°C	21.8	Almarsdottir

4. Conclusions

Hydration treatment had a positive significant effect ($p<0.001$) on the biodegradability of wheat straw measured as hydrogen production. The highest hydrogen production was reached by using a biological effluent as hydration media. Regarding the particle size, the hydrogen production was higher at 3mm than that at 0.2mm mainly for untreated wheat straw. The hydration of lignocellulosic substrates represents a key potential to be utilized for the production of biofuels at low cost since the yields can be increased up to five times than those with raw substrates. Therefore, it is important the study of hydration properties of other substrates in order to obtain the information required. Additionally, another hydration media such as surface-modified agents can be studied for this purpose.

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