Gaseous Biofuels Production from Sweet Sorghum and Olive Pulp

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Abstract

Biomass from energy crops and agroindustrial wastes can be biologically converted to liquid or gaseous fuels, such as ethanol, methanol, methane and hydrogen, which was recently characterized as the fuel of the future. Hydrogen is a clean and environmentally friendly fuel, which produces water instead of greenhouse gases when combusted. It can be produced by renewable raw materials, such as organic wastes, and possesses a high-energy yield (122 kJ/g) due to its light weight. Furthermore, hydrogen could be directly used to produce electricity through fuel cells. Biological hydrogen production, one of the several ways to produce hydrogen, has received special attention during the last decade.

Biohydrogen may be produced by cyanobacteria and algae through biophotolysis of water, or by photosynthetic and chemosynthetic - fermentative bacteria. Anaerobic fermentative bacteria produce hydrogen without photoenergy, and so the cost of hydrogen production is 340 times lower than the photosynthetic process. In addition, carbohydrates are the main source of hydrogen during fermentative processes and therefore wastes/wastewater or agricultural residues rich in carbohydrates can be considered as potential sources of hydrogen. The hydrogen yield varies depending on the final metabolic products, mainly volatile fatty acids (acetic, propionic and butyric acids), lactic acid and ethanol.

The effluent from a hydrogen producing reactor may be subjected to a subsequent anaerobic digestion step with the conversion of the remaining organic content to biogas (mainly methane and carbon dioxide), which may also be used as a fuel for the production of electricity.

In this study, a two-step continuous process is developed for the biological hydrogen production and the subsequent production of biogas from energy crops and agroindustrial wastes. The process feasibility is examined for the cases of (a) sweet sorghum and (b) olive pulp, as feed materials.

Sweet sorghum is rich in readily fermentable sugars and thus it can be considered as an excellent raw material for fermentative hydrogen production. The fermentative production of hydrogen from the sugars contained in a sorghum extract in a continuous stirred tank type bioreactor was examined at various hydraulic retention times. The effluent from this reactor was subsequently fed to a continuous stirred tank type anaerobic digester for the production of methane. Furthermore, the methane potential of the solids remaining after the extraction process was determined and the overall potential of sweet sorghum biomass for hydrogen and methane production was assessed.

It was shown that continuous fermentative hydrogen production from sweet sorghum extract is possible and stable using the indigenous microflora without a pre-heating step. The highest biogas and hydrogen production rate was obtained at the HRT of 6h while the highest yield of hydrogen produced per kg of sorghum biomass was achieved at the HRT of 12h.

The second application considered was the production of gaseous fuels from olive-pulp, a semi-solid residue generated from two phase olive-mills. The

replacement of three-phase olive mills by their two-phase counterparts is a very promising perspective from an environmental point of view, but its feasibility depends on the ability to exploit the generated olive-pulp. Again it was shown that a two-step process based on biohydrogen production followed by production of biogas is indeed stable and feasible. The performance of the process for the two different substrates is compared and discussed.

Moreover, the present study showed that sweet sorghum extract could be used for hydrogen and methane production in a two-stage process. It was proved that the effluent from the hydrogenogenic reactor is an ideal substrate for methane production.

This work demonstrated that biohydrogen production can be very efficiently coupled with a subsequent step of methane production and that biomass from energy crops and agroindustrial wastes could be ideal substrates for a combined gaseous biofuels production.

1. Introduction

In the early stages of human activity energy use (heating, cooking etc) was based on biomass (wood). With the invention of the steam engine (1781), coal was used as fuel. Coal was later complemented with the use of petroleum and natural gas, all with high carbon content.

It is well known that the carbon dioxide which is produced from burning fossil fuels such as coal, oil and natural gas is responsible for the greenhouse effect. In the last century the population of earth has increased by a factor of 6, while the consumption of energy has increased by a factor of 80. This astonishing rise in energy demands, coupled with the environmental consequences of fossil fuel usage and the anticipated shortage of such sources in the future has forced modern society to turn vividly to renewable energy sources.

Biomass is all material of biological origin (e.g. energy crops, animal, industrial and agricultural waste). On the average it consists of 25% lignin and 75% sugars, after subtracting an additional 5% of other compounds. Today it covers 14% of total energy needs, but only 38% of the energy potential of biomass is actually utilized (International Energy Agency, 1998).

Biomass may be utilized for energy production either through thermal (incineration, pyrolysis, gasification) or through biological processes (Demirbas, 2004). The biotechnological exploitation of biomass is an anaerobic process with no environmental impacts. It produces added-value fuels such as (Claasen et al., 1999):

- bioethanol (a mature technology)
- biogas (anaerobic digestion, a mature technology)
- Hydrogen (a developing technology)
- Microbial fuel cells (just starting)

Historically, we have observed an evolution from high carbon content fuels to high hydrogen content fuels and from solid fuels to liquid and gaseous fuels, as shown on Fig.1. Today hydrogen is considered as the energy carrier of the future.

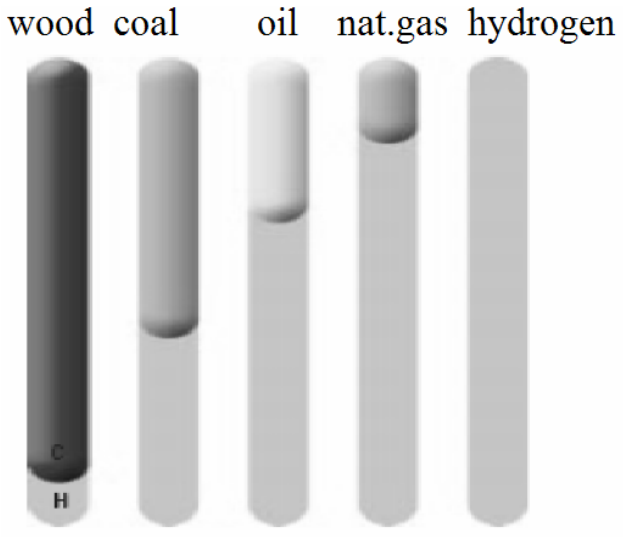

Fig.1. Carbon and hydrogen content of various fuels.

Anaerobic digestion is the process of conversion of organics to CH_4 and CO_2 (biogas), through the concerted action of a mixture of microbes in the absence of oxygen. The overall reaction may be written as:

Organic matter + water → $CH_4 + CO_2 + NH_3 + H_2S$ + new cells + heat

The generated biogas, depending on the degree of reduction of the feed substrate has a typical composition of 50-70% CH_4 and 30-50% CO_2. The energy yield of biogas, depending on its composition varies in the range 18.700-26.000 kJ/m^3 (35.800 kJ/m^3 for pure methane). It is apparent that anaerobic digestion may be used to produce biogas out of various organic sources such as solid waste, agroindutrial wastewaters, energy crops etc (Gavala et al., 1999; Gavala et al., 2004, Brian et al., 1991).

Hydrogen as already discussed, is a very attractive fuel since it may be used in fuel cells (Lay et al., 1999; Benemann, 1996). These are devices with zero pollutant emissions that are anticipated to replace internal combustion engines in the future. Vehicles powered through fuel cells have already been constructed and are now in the process of being optimised.

Hydrogen, just like biomass, may be produced from renewable sources, such as energy crops and organic waste (Kapdan and Kargi, 2006). In addition it may be produced by water (Asada and Miyaka, 1999). It has a very high energy (122 kJ/g) yield in comparison with methane or ethanol and its burning does not contribute to the greenhouse effect, as it produces water as the sole product.

Hydrogen may be produced from (a) conventional resources such as natural gas (through steam reforming or thermal breakdown), (b) water through photolysis or electrolysis, and (c) biomass through pyrolysis, gasification or biological production. From Table 1 it becomes apparent that biological production of hydrogen combines low cost with no CO_2 emissions, and is therefore the method of choice.

Table 1 .Cost and emissions of alternative hydrogen production methods

Technology	**Cost (€ / m^3 H_2)**	**Emissions of CO_2**
Steam generator of natural gas	32	0.8
Conventional electrolysis	23	1.8
Electrolysis with CO_2 Dependent electricity	27-36	0
Biotechnological production of H_2 from biomass	21	0
Steam generation from biogas	32	0
Electrolysis using wind energy	25	0
Electrolysis using photovoltaic cells	295	0

Biotechnological methods that have been developed for hydrogen production include:

1. water biophotolysis from algae and cyanobacteria
2. Photodegradation of organics by photosynthetic bacteria
3. Fermentative hydrogen production from organic matter (degradation of carbohydrates, volatile fatty acids, alcohols, H_2)
4. Hybrid systems of photosynthetic and fermentative bacteria

We consider the fermentative hydrogen production as the most promising method at this stage. Glucose biodegradation follows one or more of the metabolic pathways presented in Fig.2

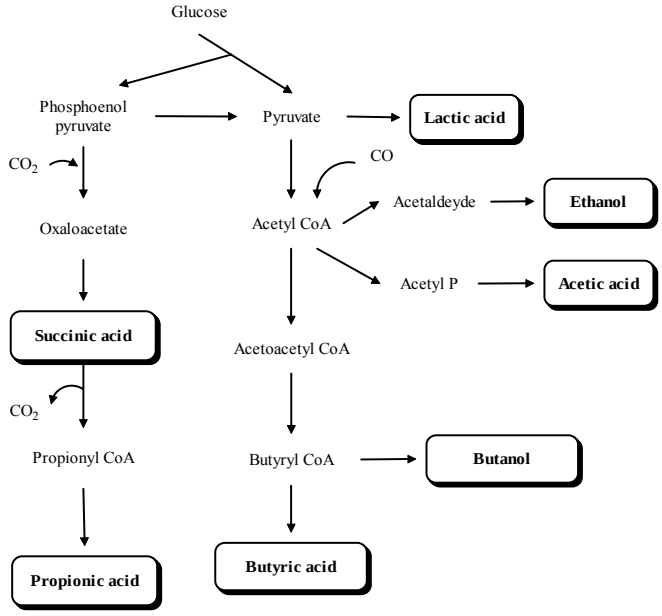

Fig.2. Anaerobic metabolic pathways of glucose.

Depending on the products, we may have a positive, negative or zero production of hydrogen. It is thus to our interest to turn the microbial metabolism to those end products, whose production is accompanied by the highest possible amounts

of hydrogen. Acetate and butyrate are such products, while lactate and ethanol are not accompanied by hydrogen production. From the reactions:

$C_6H_{12}O_6 + 2H_2O \rightarrow 2CH_3COOH + 2CO_2 + 4H_2$
$C_6H_{12}O_6 \rightarrow CH_3CH_2CH_2COOH + 2CO_2 + 2H_2$
it becomes apparent that acetate production is accompanied by 4 moles of hydrogen per mole of glucose, while butyrate production is accompanied by only 2 (Nandi and Sengupta, 1998; Hawkes et al., 2002). Thus, a yield between 2 and 4 is expected, if we have a mixture of the two acids being produced (Chen et al., 2001a; Mizuno et al., 2000; Fang and Liu, 2002).

The production of H_2 in a continuous bioreactor will depend on factors such as the pH (typically low), the Hydraulic Retention Time (typically small enough to exclude methanogens), the temperature, the concentration of nutrients, the stirring rate and the organic loading to the reactor (Lay, 2000; Ueno et al, 1996; Lee et al., 2002; Zoetemeyer et al., 1982; Ren et al. 1997). The effluent from a hydrogen producing reactor contains high concentrations of fatty acids. It may be thus fed to an anaerobic digester producing methane as a secondary product. The effluent from the digester is sufficiently stabilized and may be used as soil amendment. In the sequel, we present our results on hydrogen and methane production from energy crops and waste.

2. Biotechnological production of hydrogen and methane from sweet sorghum

Sweet sorghum biomass is rich in readily fermentable sugars (Table 2) and thus it can be considered as an excellent raw material for fermentative hydrogen production. Our concept for complete exploitation of sweet sorghum for the production of hydrogen and methane is shown on Fig.3. The process is a two step process that involves a) the fermentative production of hydrogen from the sugars contained in a sorghum extract in a continuous stirred tank type bioreactor (H_2-CSTR) at various hydraulic retention times, and b) the subsequent anaerobic treatment of the effluent of the H_2-CSTR with the simultaneous production of methane in a continuous stirred tank type reactor as well (CH_4-CSTR). Furthermore, the methane potential of the solids remaining after the extraction process is determined and the overall potential of sweet sorghum biomass for hydrogen and methane production is assessed.
Table 2. Composition of sweet sorghum.

Type of carbohydrate	% dry weight
Sucrose	55
Glucose	3.2
Cellulose	12.4
Hemicellulose	10.2

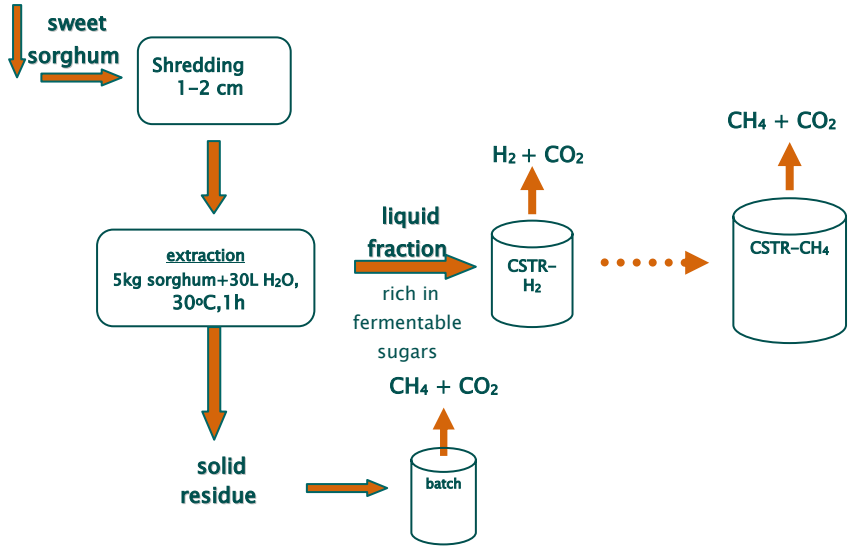

Fig.3 Fuels from sweet sorghum

Sweet sorghum var. Keller seeds were sown at mid of May in the University of Patras experimental station and the stalks were harvested in mid October. The stems were stripped from the leaves, chopped to a size of 20 cm and stored in the freezer at –20ºC. Subsequently, the stalks were milled to an average particle size of 1-2 mm. Extraction of free sugars of the sorghum biomass was done in batches, by mixing 5 kg of milled sorghum stalks with 30 L of tap water for 1 h, at 30ºC. After the extraction process, a liquid fraction (sorghum extract), rich in soluble carbohydrates and a solid fraction (sorghum cellulosic-hemicellulosic residues) were obtained. Sorghum extract and residues were used as substrate for hydrogen production by mixed acidogenic cultures. Table 3 presents the main characteristics of the sorghum extract.

Table 3. The main characteristics of sorghum extract.

Characteristics	Value
pH	7.5 ± 0.5
TSS, g/l	1.98 ± 0.27
VSS, g/l	1.87 ± 0.35
Total COD, g/l	18.5 ± 2.5
Soluble COD, g/l	17.5 ± 2.0
Soluble carbohydrates, g/l	17.0 ± 2.0
Total Kjendhal nitrogen, g/l	0.025
Total phosphorus, g/l	0.035

Continuous experiments for biohydrogen production

A 500 ml active volume mesophilic (35°C) CSTR-type digester (H_2-CSTR) was started-up and fed with sorghum extract. The reactor was operated anaerobically at hydraulic retention times (HRT) of 24, 12, 8, 6 and 4 h. Hydrogen production as a function at steady-state for various HRTs is shown on Table 4. It becomes apparent that the highest production is achieved for an HRT of 6 hrs. Although the productivity of hydrogen is maximum for HRT =6h, the highest stoichiometric yield of hydrogen

is obtained for HRT =12h and is 0,9 mol H_2/ mol converted glucose, corresponding to a hydrogen production of 10,4 L H_2/ kg sorghum.

Table 4. Hydrogen production for various HRTs.

HRT (h)	mmolH_2/d	mol H_2/ mol glucose	L H_2/ Kg sorghum
24	15.9	0.37 ± 0.02	4.9
12	66.39	0.86 ± 0.04	10.4
8	84.35	0.75 ± 0.05	8.4
6	103.6	0.70 ± 0.02	7.6
4	89.77	0.41 ± 0.02	4.3

Continuous experiments for methane production

A 3 l active volume mesophilic (35°C) CSTR-type digester (CH_4-CSTR) was started up using anaerobic sludge and fed with the effluent of the hydrogenogenic reactor. The digester was operated anaerobically at a hydraulic retention time of 20d. The mean values of the main characteristics of the influent of the anaerobic digester after collection of the effluent of the H_2-CSTR, homogenization and preservation at -20ºC, as it has already been mentioned, are presented in table 5. The influent of the methanogenic reactor was rich in volatile fatty acids, as it was anticipated, with the concentration of soluble carbohydrates being almost negligible compared with the soluble COD concentration.

Table 5.The main characteristics of the influent of the CH_4-CSTR

Characteristic	Value
pH	4.7 ± 0.5
TSS, g/l	1.9 ± 0.1
VSS, g/l	1.5 ± 0.1
Soluble COD, g/l	15.9 ± 1.5
Soluble carbohydrates, g COD/l	0.9 ± 0.3
Volatile Fatty Acids, gCOD/l	12.2 ± 2.0

The digester was operated at an HRT of 20d for more than six hydraulic retention times, with an influent flow rate of 150 ml/d. The characteristics of the CH_4-CSTR at steady state are presented in table 6. The percentage of COD removal was

approximately 97%, implying that the performance of the CSTR is not kinetically limited. This, of course implies, that it should be possible to reduce somewhat the HRT, without loss in performance. The methane production rate reached 0.73 l CH_4 per day giving a yield of 4.9 l methane per l of influent. This latter corresponded to a yield of 29 l CH_4/ kg sweet sorghum, considering that 5 kg of sweet sorghum biomass were initially mixed with 30 l of water.

Table 6. The characteristics of the CH_4-CSTR at steady state.

Characteristic	Value
pH	7.5 ± 0.1
Alkalinity, mg $CaCO_3$/l	6650 ± 50
Biogas production, l/d	1.14 ± 0.12
% in CH_4	64
Soluble COD, mg/l	560 ± 170
Acetic acid, mg/l	50 ±10
Butyric acid, mg/l	40 ± 5

Batch experiments for methane potential determination

Batch experiments were carried out at 35ºC in 160 ml serum vials, in order to determine the methane potential of the solid fraction obtained after the extraction process. The calculated methane potential of the solid residues, after subtracting the control values is 6 mmol CH_4 / g TS. This yield corresponds to the production of 39 l CH_4/ kg of sweet sorghum.

3. Biotechnological production of hydrogen and methane from olive pulp

One major environmental concern in the Mediterranean countries is the generation of high organic content (COD up to 150 g/l) olive mill wastewater from the traditional, three-phase, olive oil producing industries. Wastewater treatment in small-scale olive mills is usually prohibitively expensive even though many studies exist on the physicochemical and/or biological treatment of olive mill wastewater with efficiencies of organic matter removal lying between 65 and 95 % (Angerosa F, 2000, Benitez F. J et al., 1999, Madejon E et al, 1998a. Madejon E et al, 1998b., Tsonis S.P. and Grigoropoulos S., 1988.). Recently, two-phase decanters have been employed for the extraction of olive oil without addition of water and without generation of wastewater. The semi-solid residue (olive pulp) coming from two-phase processing of olives is rich in carbohydrates and organic matter (Angerosa et al., 2000; Skiadas et al., 2004). Olive pulp could be an ideal substrate for the production of energy in the form of hydrogen and methane (Skiadas et al., 2004; Gavala et., 2005; Gavala et al., 2006, Koutrouli et al., 2006).

The characteristics of the olive-pulp that we determined are given on Table 7. To avoid problems of clogging, the olive pulp was diluted by a factor of four with tap-water.

Contnuous experiments for production of hydrogen from olive pulp

Experiments were carried out in a 500 mp CSTR operated mesophilically (35^{o}C). Retention times tested were 30 h, 14.5 h and 7.5 h. Tables 7, 8 and Fig.4 present the results.

Table 7. Olive pulp characteristics.

Parameter	Value
density (g/ml)	1.092
seeds (g/100 g olive pulp)	8.9 ± 0.2
moisture %	70.5
g TS/100 g olive pulp	29.5 ± 1.2
g VS/100 g TS	94.84 ± 0.1
Total carbohydrates (g/100 g TS)	24.5 ± 5.6
N (g/100 g TS)	2.2
P (g/100 g TS)	0.075
COD (g/100 g TS)	158 ± 10.1
lignin (g/100 g TS)	38.4

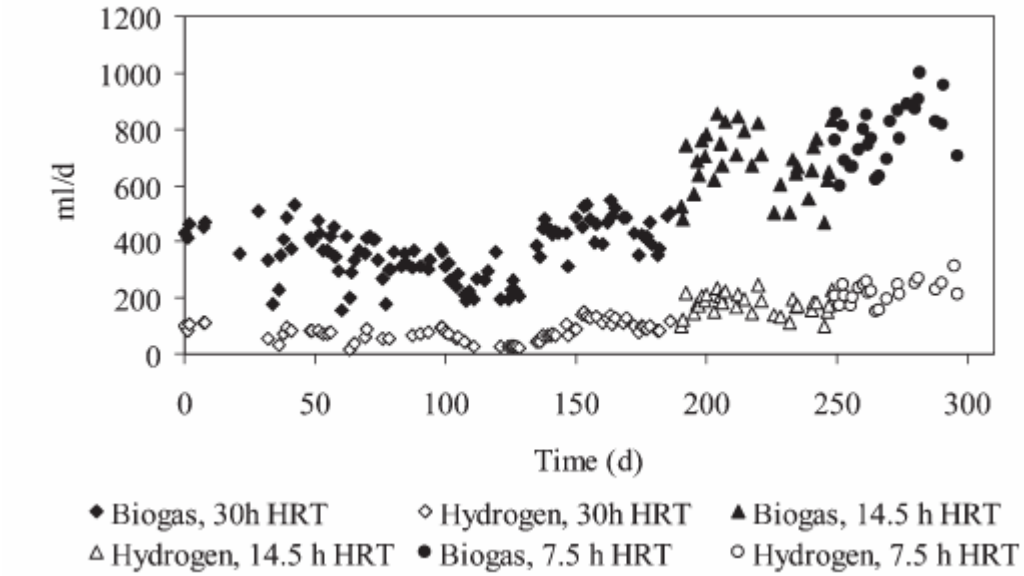

Figure 4. Biogas and hydrogen evolution during the experiments in the CSTR-type hydrogenogenic reactor.

Table 8 Reactor performance as a function of HRT in hydrogenogenic reactor receiving olive-pulp.

HRT (h)	30	14.5	7.5
pH	4.9 ± 0.1	4.8 ± 0.1	4.9 ± 0.1
% H_2	26.4 ± 1.7	26.7 ± 1.4	29.1 ± 1.6
ml produced gas / day	490 ± 53	737 ± 82	791 ± 85
ml produced H_2 / day	130 ± 4	196 ± 24	231 ± 22
mmol H_2/g soluble carb.tes consumed	4.5 ± 0.2	3.4 ± 0.6	2.8 ± 0.4
Soluble carbohydrate conversion (%)	93	69	40

We observe that as the HRT is reduced, the pH is maintained constant, there is an increase in hydrogen production, but a reduction in the stoichiometric yield. In addition there is reduction of the percentage of carbohydrates converted. Also (data not presented here) we saw a reduction of acetate, butyrate and ethanol production.

Continuous experiments for production of methane from anaerobic digestion of olive pulp and pretreated olive pulp for hydrogen production.

Two 3-Liter CSTRs were used in order to test the performance of mesophilic (35ºC) and thermophilic (55ºC) anaerobic digestion of olive pulp (diluted 1:4) ,respectively. A third reactor received the effluent of the hydrogenogenic reactor, and was also operated mesophilically (35ºC). Tables 9 and 10 present the main steady state characteristics of the first two reactors. We observe similar biogas yields at both hydraulic retention times tested. Table 11 presents the corresponding results for the digester which receives a feed that has been stripped for hydrogen. This reactor was operated at 20 d. Upon comparison with tables 9 and 10, we notice that having stripped the olive pulp for hydrogen leaves a waste that contains practically the same methanogenic potential as the original feed.

Table 9. 3L mesophilic digester receiving olive pulp– Steady states

HRT (days)	20.41 ± 1.70	10.17 ± 1.13
pH	7.52 ± 0.04	7.20 ± 0.07
VFAs (mg/l)	207 ± 66	222 ± 66
Biogas (l/l/d)	1.02 ± 0.09	2.00 ± 0.08
% CH_4	67 ± 4	65 ± 2
d.COD (g/l)	7.25 ± 0.93	7.23 ± 0.69
TSS (g/l)	31.66 ± 2.01	29.61 ± 1.16
VSS (g/l)	28.43 ± 1.77	27.51 ± 1.02

Table 10. 3L thermophilic digester receiving olive pulp– Steady states

HRT (days)	20.60 ± 2.07	10.02 ± 0.30
pH	7.53 ± 0.06	7.25 ± 0.03
VFAs (mg/l)	117 ± 49	14 ± 9
Biogas (l/l/d)	1.08 ± 0.08	1.89 ± 0.04
% CH_4	65 ± 4.5	64 ± 1.9

d.COD (g/l)	7.25 ± 0.93	7.94 ± 0.42
TSS (g/l)	29.34 ± 1.57	31.62 ± 2
VSS (g/l)	26.63 ± 1.46	29.58 ± 1.85

Table 11. Mesophilic digester fed with pretreated olive pulp – steady states

HRT (days)	20.04 ± 0.63
pH	7.62 ± 0.05
VFAs (mg/l)	488 ± 83
Biogas (l/l/d)	0.96 ± 0.05
% CH4	65.7 ± 2.4
d.COD (g/l)	8.04 ± 0.60
TSS (g/l)	43.82 ± 1.45
VSS (g/l)	39.82 ± 1.17

4. Conclusions

It has been shown that continuous fermentative hydrogen production from sweet sorghum extract is possible and stable using the indigenous microflora without a pre-heating step. The highest biogas and hydrogen production rate was obtained at the HRT of 6h while the highest yield of hydrogen produced per kg of sorghum biomass was achieved at the HRT of 12h (10.4 l H_2 / kg sweet sorghum). Moreover, the present study showed that sweet sorghum extract could be used for hydrogen and methane production in a two-stage process. It has been proved that the effluent from the hydrogenogenic reactor is an ideal substrate for methane production with approximately 70 l CH_4 / kg sweet sorghum, 39 l of which come from the solid residues. Continuous methane production of the H_2-CSTR effluent yielded 29 l CH_4 / kg of sweet sorghum, while the methanogenic potential of the solids residues after the extraction process yielded also 39 l CH_4 / kg sweet sorghum.

Olive pulp may be used for hydrogen production, although the hydrogen yiels are rather low. Perhaps they may be increased through appropriate pretreatment that would increase dissolved carbohydrates. On the other, hand it is a suitable substrate for methane production by anaerobic digestion (1 m^3 olive pulp produces 54 m^3 of methane).

To sum up, this work demonstrated that biohydrogen production can be very efficiently coupled with a subsequent step of methane production and that energy crops such as sweet sorghum, but also wastes, such as olive pulp, could be an ideal substrate for a combined gaseous biofuels production.

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