



Synthesis of Bio-Hydrogen Renovated with Carbohydrate Rich Wastewater, Utilizing Monitoring Based Agitable UASB Reactor

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Abstract:- During past one decade, there has been a resurgence of interest in biological hydrogen production. The conducted experiment have been utilizing numerous organic resources among them carbohydrate is proven to be one of the major sources, which yields a significantly larger quantity of bio-hydrogen compares to other organic resource materials. The current study is to investigate the biological hydrogen production by utilizing distillery spent wash. Pilot-scale MAUASB reactor was fabricated and operated in order to investigate the bio-hydrogen yield from distillery spent wash, maintained at a constant temperature 35°C. The study reveals H₂ gas production gradually increases up to 8th day and thereafter a decrement was observed in the level of bio-hydrogen production due to the formation of methane. Quantifying the overall bio-hydrogen production, a potential growth was significant between the 7th and 8th day of reactor startup, which shows a satisfactory value of 222.1 ml and 272.4 ml at the pH of 5.5 and 5.1 respectively. The influence of pH was proven to be the major governing factor for the biological hydrogen production. Hence the study concluded that the reactor operating conditions (acidophilic pH 5.0-5.5) were found to be favorable for effective bio-hydrogen gas production while maintaining an optimum COD removal rate of 81% at the pH value of 5.0.

Keywords: Anaerobic treatment, Bio-hydrogen fuel, Carbohydrate rich wastewater, COD removal, Monitoring Based Agitable UASB Reactor.

1. Introduction

Nowadays, universal energy necessities are mostly dependent on fossil fuel, which will ultimately lead to notable depletion due to limited fossil energy resources. It reveals that hydrogen has theoretically high energy yield of 122 kJ/g, which is 2.75 times greater than hydrocarbon [22] and it is considered to be an ultimate alternative energy source with no environmental interventions [17] producing water vapor being the only by-product [18]. In modern days, when growing energy scarcity is a hot topic and considered as a global threat, in that context a green technology like bio-hydrogen should be paid more attention [39]. Hydrogen can be produced by biological fermentation method, by the way of using microorganisms. Dark fermentation is one of the most attractive biological method compared to photo fermentation since a variety of carbon sources can be utilize as substrates and hydrogen is produced continually without depending on solar energy [9,44]. Synthesis of bio-hydrogen from organic substrate comprises exclusive advantages like; high rate of bacterial growth requires low energy input, no oxygen limitation problems, economic feasibility proclaimed by using dark fermentation process[20]. Organic wastes consumed as feedstock among molasses-based wastewater can be appropriate for bio-energy production subsequently its having high organic and inorganic content[29]. Dark fermentation using wastewater as substrates which is environmentally sustainable for hydrogen production and attractive efficiently since it allows together energy recovery and mitigation of waste [40]. Substrate concentration and pH are the key operating and environmental factors in this biological process by mixed cultures. Different metabolic pathway and microbial community structures have been experimental in mesophilic, thermophilic and extreme thermophilic environment [4, 21, 6]. Because of a number of advantages over mesophilic practice including higher yields of hydrogen, easy recovery for liquid product (ethanol), and narrow spectrum of ending fermentative products [8, 38]. Literature suggests carbon sources can be effectively used as a substrates hence it will produce bio-hydrogen all day long without light. A wide range of carbonaceous waste materials can be used as a substrate for bio-hydrogen production such as sugar wastewater, starch wastewater, and dairy wastewater. Amid the yield of bio-hydrogen is optimum from Sugar wastewater than other raw materials as it contains sucrose and glucose, which are simple sugars and can easily be converted to hydrogen at elevated temperatures with higher conversion efficiencies due to its readily biodegradable carbon source, which can be found in most of the industrial effluents. This specifies that sugar industry wastewater can be commercially used for industrial bio-hydrogen production [32]. As per the early study both pure culture such as *Clostridium* sp. and mixed cultures of anaerobic bacteria, have been used to convert carbohydrates (e.g., glucose) to H₂ [11, 42, 25, 45]. Furthermore, bio-hydrogen production by mixed cultures potentially necessitates a pre-treatment step in

order to restrain hydrogen-consuming bacterial activity, therefore increasing the process operational complexity and price. The different pre-treatment methods reported in hydrogen production processes however heat-shock pre-treatment is the most broadly applied [12, 18]. Hydrogen production is possible by continuously operating bioreactors have been investigated by several researchers [28, 48, 10, 25, 33], but still, technical lagging remains in sustainable hydrogen production. Despite hydrogen is currently accepted as an environmentally safe energy source [9]. During the fermentation of sugars, the pH of the medium is known to regulate and modify to solventogenesis [19]. Choosing appropriate pH is also an important factor for improve hydrogen production due to the effects of pH on both hydrogenase activity or metabolic pathways [46] and [9] have reported that the maximum hydrogen yield occurred at a pH value of 5.5, whereas [27] reported that the maximum hydrogen yield was achieved at an initial pH of 9.0 [14] found that the maximum hydrogen yield occurred with an initial pH of 7. These inconsistent results appear to be due to a lack of buffering capacity that would avoid the pH from declining. Since a practical point of analysis, it is important to examine in what way the initial pH control the hydrogen production when no pH control is used during fermentation.

1.1. Biological Hydrogen pathway

The anaerobic treatment is bioconversion technique of digesting organic substances and yield carbon dioxide and methane in association with the meagre bacterial growth which takes place in absence of oxygen. The digestion procedure comprises quite a few inter-reliant, multifaceted chronological and analogous biological reactions. The products from one group of microorganisms help as the substrates for the next from these reactions [34]. The overall conversion process is often described as a three-stage process namely, hydrolysis and liquefaction, acidogenesis and methane fermentation, which occurs simultaneously within the anaerobic digester [36]. The first is the hydrolysis of insoluble biodegradable organic matter, the second is the production of acid from smaller soluble organic molecules, and the third is methane generation. Before methane could be fermented, the bioconversion could be interrupted and pure hydrogen can be captured as an intermediate by-product.

The main objective of this study was to enhance the yield of the biological hydrogen from carbohydrate-rich industrial wastewater under anaerobic microenvironment, utilizing monitoring based Agitable UASB reactor. Furthermore to address the influence of pH and total solids on hydrogen synthesis efficiency and establish a correlation interface between the Bio-hydrogen production and COD removal.

2. Materials and methods

2.1. Waste characteristics

In order to conduct the experimental work, wastewater collected from sugar industry Cuddalore, Tamil Nadu, India was used as the substrate. The collected sugar industry effluent was the mixture of wastewater generated from different activities took place during the processing of sugarcane into raw sugar. The wastewater generated contains high BOD and suspended solids which need to be removed. Surplus waste was formed because of the leakages and spillage of juice, molasses, and syrup. The periodical washings of the floor also contribute a great lot to the pollution load. The industrial effluent collected to perform the present study was the combination of the effluent from utensil cleaning, plus other day to day activities related to the production site and the characteristics of the same is tabulated below (table 1). In this context, it's quite mandatory to say that the quality of the data obtained from the analysis of sugar industry wastewater samples showed slightly different characteristics compare to the reported typical quality of sugary wastewater.

Table 1: Characteristics of untreated wastewater sample

S. No.	Parameters*	Values (mg/l)
1	pH	4.0
2	Chemical Oxygen Demand (COD)	48000
3	Biochemical Oxygen Demand (BOD)	15000
4	Total Dissolved Solids (TDS)	11080
5	Total Solids (TS)	17540
6	Total Suspended Solids (TSS)	6460
7	Volatile Solids (VS)	7534

Except pH all other parameter values are expressed in mg/l

2.2. Portrayal of the bioreactor system

The schematic representation and overview of the Monitoring Based Agitable Up-flow Anaerobic Sludge Blanket Reactor have been shown in figure 1 and 2 respectively. The system comprises four automated units, including a feeding tank, main body of reactor, gas measuring sensor and automated temperature control system. The feed tank has the supply volume of 10 L and the total experimental volume of the reactor was 21 L out of which 5L volume was meant for gas collection in the

chamber located at the top of the reactor and 16 L as working volume to perform the bioconversion. The bioreactor dimensions were measured as mentioned i.e. diameter of 212 mm and height of 460 mm. The entire bioconversion mechanism took place in 4 different segments of the bioreactor namely, seed sludge introduction area at the bottom, substrate configuration part at the middle, biofilm placed in between the substrate and the gas collection chamber at the top of the reactor.

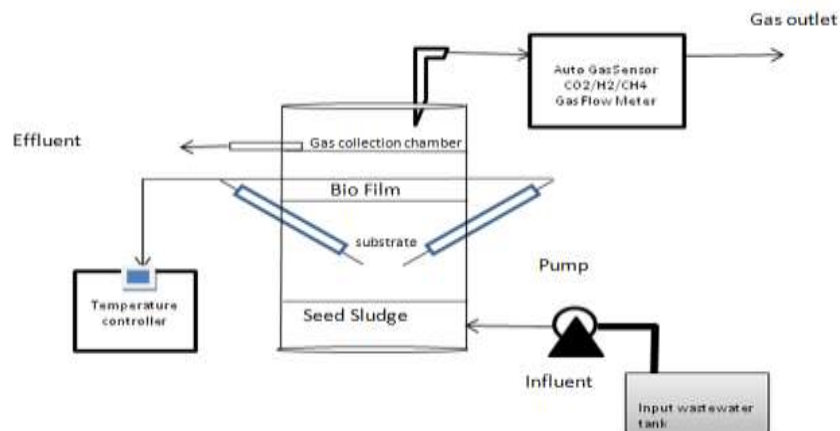


Fig. 1: Schematic diagram of MAUASB reactor



Fig. 2: An overview of the lab scale prototype MAUASB reactor

2.3. Analysis Method

Chemical analysis was performed for both influent as well as effluent. The quantity of Total Solids (TS), Volatile Solids (VS), Suspended Solids (TSS), COD and BOD5 were determined based on the standard methods (APHA, 1995). The total volume of biogas evolved was measured by a syringe and biogas composition was analyzed by a GAS Chromatography (GC 7410) equipped with a thermal conductivity detector (TCD). Stainless steel column packed with nitrogen gas was used as carrier gas for hydrogen analysis. The temperature of injector and column was maintained at 80°C.

2.4. Characterization of the seed sludge and substrate

The seed sludge was obtained from the anaerobic digester unit of sugar industry Cuddalore, Tamilnadu, India. The seed sludge was characterized as per the solid content and the results obtained were as follows, total solid of 30.7 g/l and volatile solid of 15.35g/l. Before sending it to the reactor it was preheated at 100°C for 15 minutes to inhibit the methanogenic activity. Pretreated sludge was seeded into MAUASB reactor at an HRT of 24 hours at a constant temperature of 35°C. It's mandatory to mention that the substrate was diluted to a certain fraction of the raw effluent obtained from the sugar industry, Cuddalore, Tamil Nadu to bring it to the supply strength of 0.5-5g/l.

2.5 Other technical detail

The reactor start-up period was minimized using the seed sludge obtained from an existing anaerobic sludge digester which operates at the inlet of the sugar industry effluent treatment plant. The influent COD concentration of 2000 mg/l was supplied

as initial substrate strength in the bioreactor at a flow rate of 14ml/min at 35°C with an HRT value of 24h. To enhance the biomass concentration, the bioreactor was operated for two days to stabilize the hydrogen gas production in the MAUASB reactor. The value of pH was maintained at 5.0-5.5 with a buffer solution of NaOH and H₂SO₄.

3. Results and Discussion

Previous studies from batch experimentation reported that the optimum initial pH for hydrogen production assorted based on different substrates, e.g., pH from 5.5 to 5.7 for sucrose [47]; [40, 13], pH of 6.5 for xylose [3] and for starch [36] and for cheese whey[31] pH of 6.0. The early investigations in continuous process stated that a pH of 5.5 for glucose [16], for starch pH of 5.2 [26], and for beer processing wastes pH of 5.8 [24] were optimum for hydrogen production. These studies imply that vaguely acidic pH from 5.1 to 5.5 assist to enhance the fermentative hydrogen production. However, our assessment, in which the pH of 5.1 was optimal for hydrogen-producing bacteria in the MAUASB, was different from these previous reports. Fermentative hydrogen production was feasible at even lesser controlled pH without decline based on our investigation. [15]Reported that the initial pH of 4.5 was the optimal condition for hydrogen fermentation from rice slurry. Operating the MAUASB at pH of 5.1 is probably more advantageous in terms of the inhibition of hydrogen consumers.

The yield of bio-hydrogen and metabolic pathway gets affected by the important environmental factor pH. The optimum pH range to attain the maximum hydrogen yield or specific hydrogen production rate was found between 5.2 and 6.0 from the conventional studies through pure or mixed cultures of bacteria [35, 36]. The effect of pH values ranging from 4.7 to 5.9 on H₂ production [3] and hydrogen production from liquid swine manure addition with glucose by mixed cultures in an anaerobic sequencing batch reactor illustrated that pH 5 as optimal at 37°C with a hydrogen yield of 1.48 L-H₂/L. In mixed culture on sugar and starch substrates the highest specific hydrogen production at a pH between 5.5 and 5.7[40]. The optimum pH 5-6 found to be favorable for hydrogen production from the above results under an anaerobic environment.

This study exemplify that the hydrogen production at pH 5.2 and 5.1 found to be 250.1ml and 272.4ml at 7th and 8th day of reactor operation which is higher as compared to the values reported by [24] showed that pH 5.5-6.0 as the optimum for peak H₂ production of 129 ml and HY of 107.5 mL-H₂/g COD with average hydrogen content of 41-44% at 35°C. The yield of bio-hydrogen and removal efficiency of COD from the waste stream depends upon various factors moreover both the phenomenon was maximized in this study by finalizing the most favorable operational conditions based on trial and error. The influence of different parameters and their impact has been signified in this section.

3.1. Influence of different operating parameters on production of bio-hydrogen

The impact of different influencing parameters like time, temperature and pH was observed during the operational period of the reactor and tabulated in table 2.

Table 2: Changes observed in monitoring parameters during the study period

Time in days	Temperature °C	pH	Hydrogen Gas obtained in ml	COD removal (%)
1	35	5.5	103.6	7
2	35	5.5	118.5	9
3	35	5.4	132.3	12
4	35	5.3	158.8	20
5	35	5.3	189.2	28
6	35	5.2	203.8	36
7	35	5.2	250.1	52
8	35	5.1	272.4	60
9	35	5.1	172.5	65
10	35	5.1	120.6	69
11	35	5.0	103.2	70
12	35	5.0	95.3	73
13	35	5.0	65.2	77
14	35	5.0	58.2	81

Since there was a high production in volatile fatty acids and a high rate of methanogenic activity, the hydrogen gas production was low in the reactor start-up period. After 48h of initiation period, the production of bio-hydrogen gas was found to be around 222.1ml, after that the bio-hydrogen gas production was gradually increased and it reaches the maximum quantity of 272.4 ml on the 8th day of reactor operation, showed in figure 3.

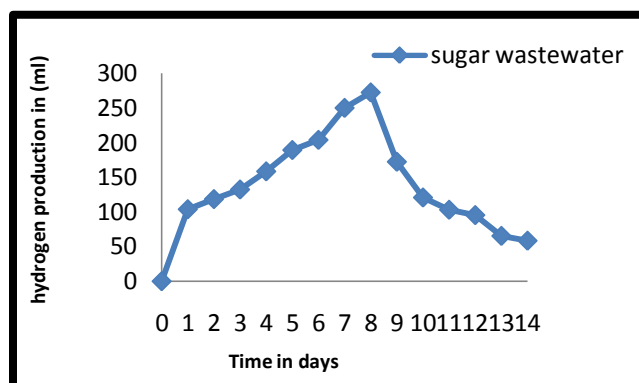


Fig.3: Correlation of time and bio-hydrogen yield from sugar industry wastewater

3.2. Correlation between COD concentrations and Bio-hydrogen production

COD reduction and formation of bio-hydrogen formulate an inversely proportional correlation up to the 8th day of reactor start-up period and thereafter albeit the change in COD level followed the similar regression, a drastic change has been observed in yield of hydrogen gas due to methane formation (Figure 4), despite there was no methane detected up to the 8th day in any of the gas collected within all hydrogen producing phase trials. A measurement study was conducted for the chemically oxidizable organic compounds present in the substrates to analyze the amount of COD removed during each process. The COD was measured for the influent and effluent for each of the experiments performed. The removal percentage was computed to understand the overall removal of COD within the trials. The hydrogen production trials reported varying degrees of removal for each of the substrates analyzed. Since the synthetic wastewater was a glucose-based substrate and almost completely soluble so it was expected to have the highest removal efficiency.

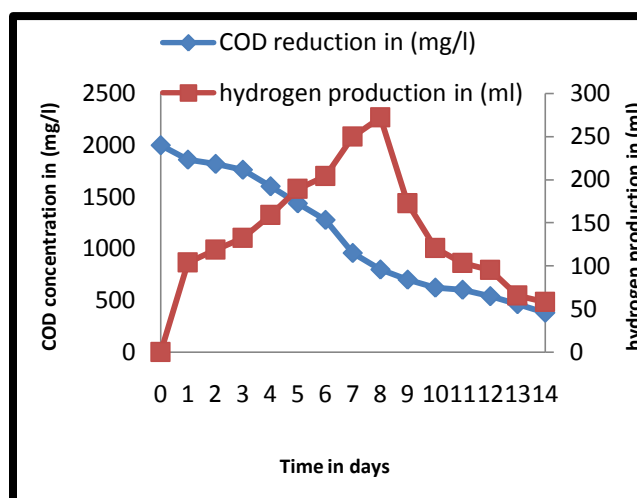


Fig.4: COD concentrations reduction Vs hydrogen production

4. Conclusion

The study claims the feasibility of bio-hydrogen synthesis from sugar industry wastewater using UASB reactor at the same time it minimizes the environmental intervention by the removal of pollution load to the optimal in sugary wastewater, due to the high content of simple sugars and prevents further nuisance. The functioning conditions (acidophilic pH 5.1) were found to be favorable for effective bio hydrogen synthesis and the maximum COD removal efficiency was found to be 81% at pH 5.0. The experimental results have shown that the maximum H₂ production (about 272.4ml) of the UASB reactor was found on 8th day maintained at pH value of 5.1 and the successive production faced depletion due to methanization at the substrate temperature of 35°C. The described process has a dual advantage of bio-hydrogen gas production with the simultaneous reduction of pollution loads of sugary wastewater in a proficient and gainful way.

5. Reference

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