ABSTRACT:

The study of influence of various wastewaters is attempted on biohydrogen production by anaerobic fermentation using periodic discontinuous suspended growth reactor. Hydrogen production through anaerobic fermentation of synthetic feed was then studied in an up flow suspended film batch reactor. Synthetic feed consists of specific concentrations of several nutrients required for anaerobic fermentation. The process parameters were set depending on the optimization studies. This process aimed at establishing hydrogen production in one liter suspended reactor. Similar studies were performed in a suspended growth anaerobic system (stirred tank reactor) having an in-built turbine and operated by a magnetic stirrer. The hydrogen production was monitored and sequencing results were used to estimate the kinetic parameters of the reaction. The suspended growth anaerobic system was fed with optimized substrate, co-substrate and nitrogen sources along with several other nutrients, which is referred to as complex feed. This process aims at studying the variations of hydrogen production with nutrient addition. Substrate conversion efficiencies of the complex feed was studied and compared with that of the synthetic feed studied in the previous stages, to establish the degree of success of the optimization process.

Keywords: Organic contents, Inorganic contents, PH, COD, VFA, HPLC, Synthetic feed, complex feed, Industrial waste Water, Enterobacter aerogenes, Clostridium sp.

INTRODUCTION

Hydrogen is one of the more suitable energy carriers for the technological and environmental prospective of the 21st century, particularly within the context of sustainable development. Hydrogen has several desirable characteristics: It has high conversion efficiency, it is recyclable and non-polluting, and yields only water after combustion. These characteristics make hydrogen the fuel of the future. The ultimate goal for conversion to the Hydrogen era is the substitution of clean hydrogen for the present fossil fuels [1]. Works on Biohydrogen production have been in progress for more than 3 decades demonstrated that a nitrogen fixing cyanobacterium, Anabaena cylindria produces hydrogen and oxygen gas simultaneously in an argon atmosphere for several hours [2, 3]. There is a practice of combined use of photosynthetic and anaerobic bacteria for the conversion of argonic acids to hydrogen. The anaerobic digestion degrades the organic matter completely to yield methane and
carbon dioxide [4]. However, the process can be interrupted by suppression of methanogenic bacteria and activation of acetogenic bacteria to produce hydrogen.

Various biomass types have been worked out as the source of hydrogen. Biohydrogen generation from the industrial effluents has not been extensively studied. Fermentative hydrogen production can be maximized with hydrogenase enzyme [5]. Decreased pH also enables suppression of hydrogen consuming bacteria and lowering of retention time encourages hydrogen production [6]. The new millennium seems to have attracted many workers to try hydrogen production from industrial wastes and also different synthetic feed combinations and conversion efficiencies) . Hydrogen production by microorganism is divided into two main categories; production by Algae or phototrophic bacteria and production by anaerobic fermentation bacteria[7]. The anaerobic hydrogen fermentating bacteria can produce hydrogen continuously without the need of photo energy[8]. Another attraction of anaerobic bio hydrogen production is that highly concentrated industrial waste water or organic waste water such as municipal solid wastes, sewage sludge can be used as raw material, which can solve pollution as well as generate hydrogen[9]. Currently anaerobic hydrogen production has been mainly studied on bench scale using pure culture such as clostridium sp, Enterobacter aerogenes the optimal condition for hydrogen production has not at been fully understood[10]. Therefore this study had three objectives (1) To study the hydrogen yielding potential of the effluents from bulk drug industry, (2) to study the biohydrogen production by anaerobic process using synthetic feed and (3) to study the relationship between the hydrogen production and quantity of the synthetic feed with different effluent concentrations. Maximum hydrogen yield mmol per day, organic loading rate were examined in the reactor kg COD / cum-day in the reactor.

**MATERIALS AND METHODS**

**Analytical Methods:** The performance of reactor with complex chemical effluents was assessed by monitoring carbon removal Chemical Oxygen Demand (COD) throughout the reactor operations and during the cycle period. In addition, pH, Oxidation-Reduction Potential (ORP), Volatile Fatty Acids(VFA), Alkalinity and Suspended Solids (SS) were determined during reactor operation to assess the performance of the reactor. The analytical procedures for monitoring the above parameters were adopted from the procedure outline in the Standard methods. The method performed for determination of physicochemical parameters was adopted from standard methods of American public health association [14]

Water used in all experiments was laboratory-distilled water (pH 6.7-7.1 and conductivity 2.5-4.3 μmho/cm) unless specified. Tap water obtained from underground source was used (color- Nil, Chlorine- Nil, turbidity- Nil, pH- 7.2-7.8, alkalinity- 40-120 mg/l, chlorides- 20-30mg/l). Innoculum development for hydrogen production, Properly pretreated mixed anaerobic sludge for process startup was procured from a lab scale UASB reactor used in treating chemical wastewater for almost 3 years. The innoculum was subjected to heat treatment at 80°C for 24 hours followed by acid treatment at pH 3 adjusted with ortho-phosphoric acid and left undisturbed for 48 hours. Further treatment with 0.2 g/l of 2-bromethansulfonic acid sodium salt (C2H4BeNaO3S) for 24 hours was performed to inhibit the methanogenic bacteria present in sludge under aseptic anaerobic conditions. A 20ml of anaerobic Innoculum was added to the anaerobic reactor in aseptic anaerobic conditions to a mixture of 33ml effluent and 22ml of

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sewage. The characteristics of the anaerobic inoculum was as followsa. Suspended Solids - 13,500 mg/lb. Volatile Suspended Solids - 7600 mg/l pH (1:1 dilutions)- 6.85.

**Reactor start up:** The reactor was inoculated with biomass acquired from an operating laboratory scale Up Flow Anaerobic Sludge Blanket Reactor (UASBR) unit, which has been in operation continuously for 3 years for the treatment of complex chemical effluents. About 300ml of the anaerobic sludge Volatile suspended solids (VSS): 3.5 g/liters) from the anaerobic reactor was acquired and fed to the suspended reactor. It was subjected to acid treatment at pH 3 adjusted with ortho-phosphoric acid and left undisturbed for 48 hours. Further treatment with 0.2 g/l of 2-bromethansulfonic acid sodium salt (C₂H₄BeNaO₃S) for 24 hours was performed to inhibit the methanogenic bacteria present in sludge under aseptic anaerobic conditions.

**Reactor operation:** The reactor has a total working volume of 1.3-liter capacity. The hydrogen fermentation was conducted at mesophilic temperature (29 ± 2°C). The pH was maintained at 6 to ensure that the fermentation process does not yield a drastic drop in the pH value after a HRT of 24 hours. This decision was based on the optimization studies. The suspended reactor was started with synthetic feed, which has the composition as shown in Table 1. About 1 liter synthetic feed was taken and fed to the reactor and the inlet and outlet samples (after a HRT of 24 hours) were collected and was continuously examined for pH, ORP, VFA, COD and hydrogen gas production. The suspension was maintained by recirculating the feed through a tube aided by a peristaltic pump operating at 100 rpm. Initial 5 days of operation in up flow feed recirculation mode produced negative results due to absence of suspension. The next 10 days operation was performed by up flow of sludge through the recirculation tube to keep the reactor in suspension. Then, sequencing was done at the HRT intervals of 1, 2, 4, 6, 8, 10, 12, 24 and 48 Hours of incubation. The samples were regularly monitored for pH, VFA, Alkalinity, COD, Glucose, Protein and Hydrogen gas parameters. High Power Liquid Chromatography (HPLC) for the samples was carried out.

**Table 1: Synthetic feed composition**

<table>
<thead>
<tr>
<th>Nutrients</th>
<th>Composition (g/l)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NH₄Cl</td>
<td>0.5</td>
</tr>
<tr>
<td>KH₂PO₄</td>
<td>0.25</td>
</tr>
<tr>
<td>K₂HPO₄</td>
<td>0.25</td>
</tr>
<tr>
<td>MgCl₂·6H₂O</td>
<td>0.3</td>
</tr>
<tr>
<td>FeCl₃</td>
<td>0.025</td>
</tr>
<tr>
<td>NiSO₄</td>
<td>0.016</td>
</tr>
<tr>
<td>CoCl₂</td>
<td>0.025</td>
</tr>
<tr>
<td>ZnCl₂</td>
<td>0.0115</td>
</tr>
<tr>
<td>CuCl₂</td>
<td>0.0105</td>
</tr>
<tr>
<td>CaCl₂</td>
<td>0.005</td>
</tr>
<tr>
<td>MnCl₂</td>
<td>0.015</td>
</tr>
<tr>
<td>Glucose (C₆H₁₂O₆)</td>
<td>3</td>
</tr>
</tbody>
</table>

The inoculum from the suspended reactor was directly transferred to a stirred tank reactor fitted with a 2-blade axial turbine consisting of a magnetic pellet that can be operated with the help of a magnetic stirrer. This reactor maintained a suspension by the movement of the turbine blades, which stirred the microbial culture to move in the working volume in an irregular manner.
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**Reactor Configuration**: The stirred tank reactor, manufactured by Nalgene, consists of a plastic vessel with a curved bottom. The reactor has a magnetic pellet at the center of a 2 axial blade turbine, which rotates about its axis with the help of magnetic force developed by a magnetic stirrer. The reactor has two openings at the top for inlet and outlet purposes. The various design details of the reactor are: Total Capacity: 2.2 liters, Working Capacity: 1 liter, Overall height: 266 mm, Outer Diameter of the reactor: 137 mm.

**Fig.1: Anaerobic batch stirred tank Reactor**

**Reactor setup and inlet conditions**: The reactor has a total working volume of 1.25-liter capacity. The hydrogen fermentation was conducted at mesophilic temperature (29 ± 2°C). The pH was maintained at 6 to ensure that the fermentation process does not yield a drastic drop in the pH value after a HRT of 24 hours. This decision was based on the optimization studies. The suspension was maintained by the movement of turbine blades powered by a magnetic stirrer operating at 100 rpm.

**Synthetic feed studies (Reactor operation)**: The reactor was started with synthetic feed, which has the composition as shown in Table 1. About 1 liter of synthetic feed was taken and fed to the reactor and the inlet and outlet samples (after a HRT of 24 hours) were collected and was continuously examined for pH, ORP, VFA, COD and hydrogen gas production. The reactor was analyzed for the various important process parameters for the inlet and outlet samples for around 13 days. After a steady state was attained, the sequencing at the HRT intervals of 1, 2, 4, 6, 8, 10, 12, 24 and 48 Hours of incubation. The samples were regularly monitored for pH, VFA, Alkalinity, COD, Glucose, VSS and Hydrogen gas parameters. HPLC for the samples was carried out. The reactor kinetics and substrate conversion efficiency was also calculated using the biomass and substrate concentrations at various time intervals in sequencing period.

**Complex Feed (Reactor operation)**: Complex feed refers to the variable concentrations of nutrients required to enhance fermentation and hydrogen production process. Based on optimization studies, the complex feed was specified as shown in the Table 2. The sucrose concentration was calculated to maintain an organic loading rate of approximately 5000 mg/l. DAP concentration was based on N: P ratio of 5:1. About 1 liter of the feed was taken and fed to the reactor and the inlet and outlet samples (after a HRT of 24 hours) were collected and was continuously examined for pH, ORP, VFA, COD and hydrogen gas production. The reactor was analyzed for the various important process parameters for the inlet and outlet samples for around 3 days. After a steady state was attained, the sequencing at the HRT intervals of 1, 2, 4, 6, 8, 10, 12, 24 and 48 Hours of incubation. The
sequencing samples were monitored for pH, VFA, Alkalinity, COD, Sucrose and Hydrogen gas parameters. HPLC for the samples was carried out. The substrate conversion efficiency was also calculated at various time intervals in sequencing period.

Table 2; Complex feed composition

<table>
<thead>
<tr>
<th>Nutrients</th>
<th>Composition (g/l)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Di- Ammonium Phosphate</td>
<td>0.5</td>
</tr>
<tr>
<td>MgCl₂.6H₂O</td>
<td>0.3</td>
</tr>
<tr>
<td>FeCl₃</td>
<td>0.025</td>
</tr>
<tr>
<td>NiSO₄</td>
<td>0.016</td>
</tr>
<tr>
<td>CoCl₂</td>
<td>0.025</td>
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<tr>
<td>ZnCl₂</td>
<td>0.0115</td>
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<td>CuCl₂</td>
<td>0.0105</td>
</tr>
<tr>
<td>CaCl₂</td>
<td>0.005</td>
</tr>
<tr>
<td>MnCl₂</td>
<td>0.015</td>
</tr>
<tr>
<td>Sucrose (C₁₁H₂₂O₁₁)</td>
<td>3.74</td>
</tr>
</tbody>
</table>

Fermentation of complex feed with industrial effluent

Reactor operation: The sucrose concentration was calculated to maintain an organic loading rate of approximately 5000 mg/l along with the industrial effluent. About 1 liter of the feed containing 50% complex feed and 50% industrial effluent was taken and fed to the reactor and the inlet and outlet samples (after a HRT of 24 hours) were collected and was continuously examined for pH, ORP, VFA, COD and hydrogen gas production. The reactor was analyzed for the various important process parameters for the inlet and outlet samples for around 3 days. After a steady state was attained, the sequencing at the HRT intervals of 1, 2, 4, 6, 8, 10, 12, 24 and 48 Hours of incubation. The sequencing samples were monitored for pH, VFA, Alkalinity, COD, Sucrose and Hydrogen gas parameters. HPLC for the samples was carried out. The substrate conversion efficiency was also calculated at various time intervals in sequencing period.

Alkalinity

Procedure: The sample was centrifuged for 5min at a speed of 3000rpm and filtered. 100ml of the centrifuged or filtered sample or a suitably diluted sample containing less than 3 meq/L VFA were taken. The sample was titrated with 0.1N HCl to pH=3 (A ml) using a pH meter. The sample was boiled for 3min in the 250ml flask to remove the CO₂. The sample was cooled immediately for 2min and the sample was titrated with 0.1N NaOH to pH =6.5 (B ml).

VFA (mg/l) =

\[
(B \times 100) - (A + 100) \times \text{dilution factor} \times 60
\]

99.23

Alkalinity (mg/l) =

\[
(A - B) \times \text{dilution factor} \times 60.
\]

Chemical Oxygen Demand (COD)

Procedure: 1 ml of the sample was taken in 10-ml COD vials and to this 1 ml distilled water, 2ml K₂Cr₂O₇ solution and 4ml H₂SO₄ reagents were added. These vials were kept in COD block digester and refluxed for a period of 2 hours at 150°C. After refluxing was completed the samples were cooled and were transferred into 100 ml beaker. Then 2 to 3 drops of ferroin indicator was added to sample solution and titrated against 0.1 N FAS, until brick red color appeared. COD was calculated by using the following formula. Color, turbidity and silica in the concentration of 500 ppm interfere in this
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estimation. Filtration can be done to remove color and turbidity.

\[
\text{COD (mg/l)} = \frac{(B-S) \times N \times 8000}{\text{ml of sample taken}}
\]

Where, B ml is amount of FAS consumed for blank, S ml is FAS consumed for sample, N is normality of Ferrous ammonium sulphate (FAS).

**Total solids (TS)**

Total solids in mg/l = \( \frac{(W_2 - W_1) \times 1000}{\text{Volume of sample in ml}} \)

Where: \( W_1 \) = Weight of empty dish in mg

\( W_2 \) = Weight of dish + Weight of dried residue in mg

**Total suspended solids (TSS)**

Total suspended solids in mg/l = \( \frac{(W_2 - W_1) \times 1000}{\text{Volume of sample in ml}} \)

Where, \( W_1 \) = Weight of filter paper in mg just before using it for filtering sample; and

\( W_2 \) = Weight of filter paper + dried residue in mg.

**Volatile Suspended Solids (VSS)**

The residue obtained above (TSS) could be used for volatile suspended solids. The residue obtained after oven drying was ignited in muffle furnace at 550+50°C for 20 minutes. Silica crucible containing residue was taken out from the furnace. The crucible was cooled in a desiccator and weighed to a constant weight (\( W_4 \)).

\[
\text{Volatile suspended Solids (VSS)} = \frac{(W_4 - W_3) \times 1000}{\text{Volume of sample in ml}}
\]

Where, \( W_3 \) = Initial weight of crucible in mg; and \( W_4 \) = Weight of crucible + residue after ignition in mg.

**Hydrogen Gas Estimation**

Hydrogen gas produced in the reactor is estimated using a gas sensor, FMK satellite 4-20 mA version (ATMI GmbH Inc.). This equipment is a generic gas-monitoring instrument with microprocessor based electronics interfacing with std. 4 to 20 mA alarm/control systems. Target gas and measuring range depend on type of sensor chosen.

The electrochemical sensors designed for use with the FMK satellite feature an integrated data memory. When a new sensor is fitted, the instrument’s electronics will load operating parameters of the sensor into microprocessor’s memory. The current flowing through the sensor is amplified electronically, digitized and temperature compensated and resulting concentration value is given as an analog 4 to 20 mA output signal. This output signal usually displays the % volume of hydrogen in the reactor air space.

**RESULTS AND DISCUSSION**

**Kinetic Parameters Evaluation:** For glucose, which is the primary carbon substrate used in this study, 4 moles of hydrogen are expected to be produced per mole of glucose utilized, so the substrate conversion efficiency (\( \eta \)) is

\[
\eta = 100 \times \frac{P}{4 \times V \times S_0}
\]

Where, \( P \) is the moles of hydrogen produced till that time and \( V \) is the culture volume in liters [15]. The substrate conversion efficiency attained maximum value at the end of 24th hour with a value of 6.34%. The substrate utilization rate (k_s) is also calculated and listed in the Table 3. The substrate conversion efficiency of glucose assuming 4 mol of hydrogen per mole of glucose.
consumed was studied to show a maximum value of 23% using pure cultures [11-12]. However, the experiments were conducted for around 8 to 15 days. No previous experiments have been conducted to find the substrate conversion efficiency during the sequencing period of 24 hours.

Table 3: Kinetic Model Parameters

<table>
<thead>
<tr>
<th>Time (hrs)</th>
<th>Hydrogen (ml/hr)</th>
<th>VSS (g/L)</th>
<th>( \mu ) (hr(^{-1} ))</th>
<th>( k_c ) (hr(^{-1} ))</th>
<th>( k_s ) (hr(^{-1} ))</th>
<th>Y</th>
<th>( r_x ) (*) (10(^{-3}))</th>
<th>( r_{g} ) (*) 10(^{-3})</th>
<th>( \eta )</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0</td>
<td>0.8</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>0</td>
</tr>
<tr>
<td>2</td>
<td>3.15</td>
<td>1.2</td>
<td>0.2</td>
<td>0.21</td>
<td>1.16</td>
<td>1.15</td>
<td>2.52</td>
<td>7.88</td>
<td>0.91</td>
</tr>
<tr>
<td>4</td>
<td>2.64</td>
<td>5.2</td>
<td>0.47</td>
<td>0.58</td>
<td>0.81</td>
<td>0.11</td>
<td>2.11</td>
<td>0.6</td>
<td>1.51</td>
</tr>
<tr>
<td>6</td>
<td>4.08</td>
<td>5.7</td>
<td>0.33</td>
<td>0.42</td>
<td>0.66</td>
<td>0.1</td>
<td>3.26</td>
<td>0.83</td>
<td>2.34</td>
</tr>
<tr>
<td>8</td>
<td>6.51</td>
<td>8.6</td>
<td>0.3</td>
<td>0.44</td>
<td>0.49</td>
<td>0.068</td>
<td>5.21</td>
<td>0.88</td>
<td>3.73</td>
</tr>
<tr>
<td>10</td>
<td>8.93</td>
<td>9.8</td>
<td>0.25</td>
<td>0.39</td>
<td>0.46</td>
<td>0.056</td>
<td>7.14</td>
<td>0.99</td>
<td>5.12</td>
</tr>
<tr>
<td>12</td>
<td>11.1</td>
<td>10.4</td>
<td>0.21</td>
<td>0.34</td>
<td>0.24</td>
<td>0.05</td>
<td>8.88</td>
<td>1.16</td>
<td>6.34</td>
</tr>
<tr>
<td>24</td>
<td>1.84</td>
<td>27.2</td>
<td>0.15</td>
<td>0</td>
<td>0.16</td>
<td>0.02</td>
<td>1.47</td>
<td>0.07</td>
<td>6.34</td>
</tr>
</tbody>
</table>

VFA evaluation through HPLC indicated presence of acetic acid and propionic acid within the system, which could be the possible substrate for hydrogen production is shown in figure 2.

Fig. 2: VFA evaluation through HPLC indicated presence of acetic acid and propionic acid within the system for glucose.

Upscaling with anaerobic suspended growth reactor using complex feed and Industrial Effluent (1:1 and 1:3): The optimization process included the selection of ideal co-substrate (sucrose) and nitrogen source Di ammonium phosphate (DAP) to examine the feasibility of hydrogen production from industrial effluent in a 50%-50% and 25%-75% mixtures of the complex feed and the industrial effluent. The inlet pH (feed) was maintained at 6 while the outlet pH monitored after detention time showed a slight variation (4 to 5.4) throughout the reaction period as in figure 3. However, the pH was low indicating similar performance as discussed above.

Fig. 3: pH values of inlet and outlet with anaerobic suspended growth reactor operation using complex feed and Industrial Effluent.
The inlet VFA as in figure 3 varied in the range of 2000±500 mg/L while the outlet VFA concentration showed a significant variation from 1800 mg/l to 3400 mg/l. The variation in VFA was evident up to 21 day of operation, and thereafter stabilized in and around 2600 mg/l indicating the steady state condition of the reactor. Alternate decrease and increase in the VFA values during initial phase of reactor operation indicating a sort of unstability present initially in the system. The variation of alkalinity lies within the range of 1000 mg/L to 4400 mg/L, which is considered to be on higher side. However, the variation in alkalinity figures of the outlet recorded after 25 days showed stabilized values. The alkalinity values variation indicated an increase in system response to acidogenic fermentation process [13].

The variation of COD reduction (%) indicates multitude of variations as the experiment proceeds indicating perfect degradation of the organic substrate present in the culture aimed towards hydrogen production. This set of experimentation was continued till the system attained maximum hydrogen production on a steady scale and further was stepped up by increasing the industrial wastewater composition in the feed.

The hydrogen production rate was found to increase after the seventh day followed by a gradual increase till the end as shown in figure 5. However a decrease in the production till 25th day which was supported by decreased COD reduction followed by maximum hydrogen production till the end. This proves advantageous for understanding the feasibility of the effluent towards hydrogen production. Conducting a sequencing procedure, which is discussed in the later part of this section, the experiment was continued by increasing the concentration of industrial wastewater.
Sucrose concentration was estimated at different intervals to verify the performance of the reactor is shown in figure 6. As observed from the graph the residual concentrations of sucrose were found to decrease with minimum concentration remaining at the end representing complete utilization of the substrate and maximum hydrogen production at the end.

VFA evaluation through HPLC indicated presence of acetic acid within the system which could be the possible substrate for hydrogen production as in Figure 7.

Upscaling study with Anaerobic Stirred Tank Reactor using complex feed and Industrial Effluent; The optimization process included the selection of ideal co-substrate as sucrose, nitrogen source as DAP to examine the feasibility of hydrogen production from industrial effluent in a 25%-75% mixture of the feed and the effluent.

Fig.8: : PH values of inlet and outlet with Anaerobic Stirred Tank Reactor using complex feed and Industrial Effluent.
The inlet pH was maintained at 6 while the outlet pH did not show any variation in its value of 3.8 to 5.0 throughout the reaction period (Fig 8). However, the pH was low indicating similar performance as discussed above.

Fig. 9: Variation of VFA and Alkalinity during Anaerobic Stirred Tank Reactor operation using complex feed and Industrial Effluent.

The variation of COD reduction % indicates multitude of variations as the experiment proceeds indicating perfect degradation of the organic substrate present in the culture aimed towards hydrogen production (Fig 11). The hydrogen production rate was found to drastically decrease on 44th day of the experiment (Fig 10). The values then soared to maximum hydrogen production 1.42 mmol/hr on the 4th day indicating an enhanced performance of the system. This proves advantageous for understanding the feasibility of the effluent towards hydrogen production. A parallel study of the residual sucrose concentration indicated least concentrations of the compound at end indicating good hydrogen production. However there was a increase in the residual sucrose concentration indicating requirement of further time for the complete utilization of feed and attainment of highest hydrogen production. VFA evaluation through HPLC indicated presence of acetic acid within the system, which could be the possible substrate for hydrogen production (Fig 12).
Summary and conclusions;

The main focus of this experimental investigation is to study the influence of various wastewater as substrates in the production of biohydrogen in suspended growth periodically operated reactors. Two types of chemical wastewater along with designed synthetic feed were used in the process of investigation. The reactor was operated with the optimized conditions established from earlier studies in the BEEC laboratory. The study compares the results obtained in hydrogen production from synthetic and complex feeds undergoing acidogenic reaction in the stirred tank reactor. Kinetic model parameters for hydrogen production from synthetic feed also formed an objective for this study. The anaerobic suspended batch reactor was supplied with the pretreated anaerobic mixed culture and was fed with synthetic feed containing glucose as co-substrate and NH₄Cl as the nitrogen source. The experimental data was consolidated and depicted in Table 4.

Table 4: Details of hydrogen production from the experimental variations studied

<table>
<thead>
<tr>
<th>S.No</th>
<th>Industrial Wastewater</th>
<th>Organic loading rate (OLR) (Kg COD/cum-day)</th>
<th>Hydrogen Production (mmol/day)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Designed synthetic wastewater</td>
<td>3.80</td>
<td>0.486</td>
</tr>
<tr>
<td>2</td>
<td>Composite chemical wastewater-1</td>
<td>4.67</td>
<td>3.45</td>
</tr>
<tr>
<td>3</td>
<td>Composite chemical wastewater-2</td>
<td>5.90</td>
<td>2.95</td>
</tr>
<tr>
<td>4</td>
<td>Bulk drug wastewater</td>
<td>5.04</td>
<td>3.49</td>
</tr>
</tbody>
</table>

It is evident from the data that the suspended growth configuration has yielded hydrogen production without process inhibition; however the process of hydrogen production seems to dependent on the type of wastewater and organic loading rate. Though efficient research study has been affected in this field with a high degree of success in achieving the established objectives and wastewater characteristics plays a major role in determining the stability in system performance especially in case of hydrogen production through anaerobic fermentation. Extensive research was in progress in Bioengineering and Environmental Centre (BEEC) laboratory of Indian Institute of Chemical Technology (IICT), Hyderabad.
regarding bioprocess monitoring during hydrogen production, reactor configuration optimization, characterization of hydrogen producing bacteria, bio augmentation strategy, etc. in the direction of biohydrogen production from industrial wastewater using periodic discontinuous process operation.

Acknowledgements:

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References


