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# Treatability study of synthetic slaughterhouse wastewater using enhanced clarifier hybrid UASB reactor

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The present paper demonstrates feasibility of treatment of synthetically prepared slaughterhouse wastewater by Hybrid Upflow Anaerobic Sludge Blanket reactor having Enhanced Clarifier (EC-HUASB). The main aim was to estimate the capacity of such reactor for removal of Chemical Oxygen Demand (COD) under various operating conditions and thereby to optimize the system performance. The said reactor was run under the COD Loading Rates (OLR) between 2.40 and 9.60 kg COD/m<sup>3</sup>/d on account of change in Hydraulic Retention Time (HRT) and COD between (24–6) h and (1000–6000) mg/L respectively. The highest COD removal percentage was recorded as 96% for the OLR of 4.00 kg COD/m<sup>3</sup>/d. The highest biogas generation was measured as 47.65 L/d at OLR of 9.60 kg COD/m<sup>3</sup>/d. The optimum OLR was found to be 8.00 kg COD/m<sup>3</sup>/d showing COD removal percentage and biogas generation of 92% and 41.5 L/d respectively.

Keywords: Slaughterhouse wastewater, EC-HUASB system, COD loading rate, COD removal percentage, biogas generation.

# Introduction

Slaughterhouse and Meat Processing Plants (MPPs) are the major industrial sectors around the globe utilizing up to 24% of the water for successful operation<sup>1–3</sup>. Slaughterhouse wastewater (SWW) arise a significant concern in agribusiness due to the lump sum amount water used during various processes such as slaughtering, processing and cleaning the working area<sup>4</sup>. To treat the SWW effluents there are many technologies available under physical, chemical, biological treatment category. Apart from the merits and demerits of individual technology, SWW characteristics, climate conditions and regulations should also be noted<sup>1,5,6</sup>.

However, to overcome the drawbacks in the earlier technologies there are few possibilities of combining the two or more technologies to treat the SWW in a cost-effective manner. This approach adopts and optimizes merits of the various technologies to reach the high-quality effluent from slaughterhouse wastewater<sup>7–12</sup>. Anaerobic treatment is the most effective biological method to treat the high organics containing industrial wastewater on account of its compatibility in high rate organic removal as well as energy production<sup>12–16</sup>.

Bioenergy (Hydrogen and Methane) and bioresources (lactic acid) are the most dominant alternative fuel for the society in the future<sup>17,18</sup>. This kind of renewable energy can be produced by the dark microbial fermentation processes or else through the high-rate AD reactors, such as Upflow Anaerobic Sludge Blanket (UASB) system. The UASB system holds various merits such as biogas production, low requirement of land area, low sludge production and economically cheaper option compared to other treatment processes. One of the main advantages of the UASB Reactor is more than 90% removal of high COD in most of industrial wastewater<sup>19,20</sup>. The underlying basic reason behind the energy generation and high treatment efficacy of the operation is due to interaction between various microbial communities within the sludge bed. However, few studies revealed that multiple syntrophic relationships of various bacteria lead to the decomposition of the composite organic substance and simultaneous biogas production<sup>21–23</sup>.

The UASB reactors treats wastewater under the suspended growth process only and it requires three phase separators to facilitate biogas generation. The combination of anaerobic filter and the UASB reactor is called as Hybrid Upflow Anaerobic Sludge Blanket (HUASB) system<sup>24</sup>. Additionally, the wastewater from slaughterhouse contains slowly biodegradable organic compounds, exhibiting unsatisfactory treatment performance by conventional anaerobic reactors. To overcome this limitation, HUASB reactor has played a crucial role in treating slaughterhouse wastewater<sup>25-28</sup>. On account of attached biomass, the HUASB reactor was used to improve biodegradation potential of the slaughterhouse wastewater and additionally to capture high amount of biogas compared to the conventional UASB reactor<sup>29-31</sup>. Earlier studies reveal that HUASB reactor performed 90-120% better while compared to the conventional UASB reactor<sup>16,32–34</sup>. This is possibly due to the interaction of different kinds of microorganism, higher contact time between organic substance and microorganisms, higher Solid Retention Time (SRT) and pH variation along the reactor height<sup>35-38</sup>. Considering all the above characteristics, HUASB process is mostly preferred for treating slaughterhouse wastewater<sup>5,39–41</sup>. The above stated studies were related with performance evaluation with respect to COD removal efficiency as well as biogas generation, which are the main two parameters to determine the efficiency of any anaerobic reactor.

The aim of the current study is to estimate the response of EC-HUASB reactor for treating high strength synthetic wastewater under continuous mode of operation. To fulfil this objective, a synthetic wastewater was prepared, which was equivalent to the slaughterhouse wastewater in respect of organic strength. The synthetic wastewater contained Dextrose ( $C_6H_{12}O_6$ ) as the basic substrate. The response of EC-HUASB system was monitored in terms of final effluent COD under continuous mode. The response of the reactor was measured under steady state to find out the optimum COD loading rate (OLR) to the reactor.

## Materials and methods

## Materials:

The synthetic slaughterhouse wastewater was made in the laboratory predominantly employing  $C_6H_{12}O_6$ ,  $NH_4CI$  and  $KH_2PO_4$  as the source of carbon, nitrogen and phosphorus respectively. Adequate volume of micronutrients such as  $CaCl_2$ ,  $FeCl_3$ ,  $MgSO_4$  etc. were also added in synthetic carbonaceous wastewater sample to enhance the growth of microorganism both for suspended growth and attached growth<sup>42,43</sup>. The composition of synthetic wastewater sample exhibiting COD of about 10000 mg/L as mentioned in Table 1. The anaerobic sludge was taken from a conventional anaerobic digester utilizing cow dung at the Ramakrishna Mission, Narendrapur, close to Kolkata. The characterization study on the sludge exhibited pH, BOD, COD and total suspended solids as 6.9, 1580 mg/L, 2200 mg/L and 33,087 mg/L respectively. The seed sludge was put into EC-HUASB system up to 1/3rd of the effective volume and diluted synthetic wastewater was filled in the rest portion.

	Table 1. Composition of synthetic carbonaceous wastewater							
SI.	Compound	Concentration						
no.		(g/L)						
1.	C <sub>6</sub> H <sub>12</sub> O <sub>6</sub>	10.00						
2.	NH <sub>4</sub> NO <sub>3</sub>	2.85						
3.	KH <sub>2</sub> PO <sub>4</sub>	0.45						
4.	FeCl <sub>3</sub>	0.25						
5.	MgSO <sub>4</sub>	0.0225						
6.	CaCl <sub>2</sub>	0.0275						
7.	K <sub>2</sub> HPO <sub>4</sub>	0.0302						

## Reactor setup:

The laboratory scale EC-HUASB system of 13.5 L total volume was made using acrylic fibre material, in which the working volume was 10.8 L. Total 12 number of sampling ports were provided to collect the treated effluent from various points. In addition, a brass made 20 mm diameter sludge withdrawal port was also provided at the bottom. The reactor was run in the closed temperature-controlled chamber to maintain the mesophilic temperature (37±2°C) to avoid the seasonal variations to the reactor. From the bottom of the reactor and above 30 cm height, 50 numbers of bio-carriers were placed to facilitate the attached growth process within the reactor. The placing of bio-carrier packing also leads to extraction of gas from the solid and liquid composite. The total biofilm surface area was 6700 m<sup>2</sup>/m<sup>3</sup>, which was comparatively high compared with the earlier studies<sup>44</sup>. The peristaltic pump (Miclins-pp 30X brand) was employed to maintain the OLR by varying the pump flow rate. The schematic diagram of the EC-HUASB system is given in Fig. 1.

#### Analytical methods:

All the relevant parameters such as pH, alkalinity, VSS, mixed MLVSS, Biochemical Oxygen BOD, COD, phosphorous, TKN were estimated as per Standard Methods<sup>45</sup>. The biogas was measured using the gas-chromatography (GC)

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Fig. 1. Schematic representation of EC-HUASB reactor.

as per Liu *et al.*, 2018. The instrument has the Flame Ionization Detector and the capillary column. The initial temperature of column and the detector was set as 150°C and 250°C respectively. N<sub>2</sub> was employed as the carrier gas at the flow rate of 2.0 ml/min<sup>46</sup>.

## Operational conditions:

The response of the HUASB system was studied under continuous mode of operation with synthetic carbonaceous wastewater. The HRT was adjusted to 6, 8, 10, 12, 18 and 24 h using peristaltic pump. The influent soluble COD of synthetic wastewater was adjusted in accordance with the HRT. During the entire study, samples were withdrawn from the topmost outlet at different time periods. Soluble samples were analysed to estimate the COD concentration. The total biomass concentration in the EC-HUASB system was maintained between (10000–15500) mg/L. The volume of  $CH_4$  production was estimated by water displacement mechanism using two interconnected water containers.

## **Results and discussion**

## Performance study on EC-HUASB system:

With a view to determine the optimum COD loading rate in case of synthetic carbonaceous wastewater, the values of % COD removal from various experimental runs were plotted with respect to COD loading rates. Hence, it has been possible to find out the maximum COD removal percentage and the respective COD loading rate. Similarly, the biogas production from various experimental runs was plotted with respect to COD loading rates. As a result, the maximum biogas production and the corresponding COD loading rate can be determined. The higher value of these two COD loading rates has been considered as '*Optimum COD loading rate*'. In the present study, the optimum COD loading rate is observed to be 8.00 kg COD/m<sup>3</sup>/day.

During first phase of operation, synthetic slaughterhouse wastewater having COD 1000 mg/L was fed with three different OLR variations such as 2.4, 3.0, 4.0 kg COD/m<sup>3</sup>/d under the HRT of 10, 8, 6 h and the COD removal percentage was attained at 85, 75 and 69% respectively. In case of new reactor start-up phase, many researchers suggested the optimum OLR between (0.25-2) kg COD/m<sup>3</sup>/d for higher HRT values of 24-72 h and exhibiting the average COD removal of (60-70)%<sup>47-50</sup>. However, in the present work the successive improvement in COD removal percentage was observed even under higher OLR, possibly on account of well acclimated sludge. In the second phase of the operation, 2000 mg/L of COD was fed under three different OLR variations such as 4.0, 4.8, 6.0 kg COD/m<sup>3</sup>/d, which showed maximum COD removal percentage of 91, 86 and 71% respectively. The COD concentration profiles under continuous runs with HRT 6 h and 8 h are shown in Fig. 2a.

In the third phase, 3000 mg/L of COD concentration was fed under three different OLRs such as 4.0, 6.0 and 7.2 kg COD/m<sup>3</sup>/d under HRT 18, 12 and 10 h, which exhibited COD removal percentage of 95, 93 and 92% respectively. In the fourth stage, 4000 mg/L of COD concentration was set under four different OLRs like 4.0, 5.32, 8.0 and 9.60 kg COD/ m<sup>3</sup>/d for HRT of 24, 18, 12 and 10 h, which showed COD removal efficiency of 96, 92, 87 and 74%. In the fifth stage of operation, COD was raised to 5000 mg/L under two different OLRs like 6.65 and 5.0 kg COD/m<sup>3</sup>/d under HRT 24 and 18 h, which showed COD removal percentage of 74 and 81% respectively. In the last stage of the operation, 6000 mg/L of COD was fed under two different OLRs such as 8.0 and 6.0 kg COD/m<sup>3</sup>/d under HRT 24 and 18 h, which showed COD removal efficiency of 71 and 69%. The COD concentration profiles under continuous runs with 10 h HRT are shown in Fig. 2b. Similarly, the COD concentration profiles under continuous runs with 12 h HRT are shown in Fig. 2c. Finally, the COD concentration profiles under continuous runs with HRT of 18 and 24 h are presented in Fig. 2d and Fig. 2e respectively.

It is to note that the loading rate of 4.0 kg  $COD/m^3/d$  is common for initial four stages corresponding to HRT varia-





Fig. 2a. Profile of effluent COD concentration under HRT 6 and 8 h.



Fig. 2b. Profile of effluent COD concentration under HRT 10 h.



Fig. 2c. Profile of effluent COD concentration under HRT 12 h.

tion between 24 and 10 h. These continuous runs showed the final COD between (150–310) mg/L. With respect to the above HRTs, when the COD concentration was raised, COD

removal was also enhanced to (96–69)%. It follows that the same OLR was adjusted by varying influent COD concentration. Whenever the high influent COD was fed to the reactor,

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Fig. 2d. Profile of effluent COD concentration under HRT 18 h.



Fig. 2e. Profile of effluent COD concentration under HRT 24 h.

COD removal efficiency was enhanced because of more interaction between the substrate and the biomass. Similarly, the loading rate of 6.0 kg COD/m<sup>3</sup>/d was applied to the system by means of influent COD of 2000 and 3000 mg/L under varying HRT 8 and 12 h, which showed COD removal efficiency of 79 and 93% respectively. It reveals that the basic wastewater dilution ratio plays a crucial role in the COD removal. Whenever the HRT was increased under the same OLR, the COD removal percentage was increased to a great extent.

On the other hand, the loading rate of 8.0 kg COD/m<sup>3</sup>/d was considered for COD 4000 and 6000 mg/L to determine the optimum reactor efficiency with respect to the HRT 12 and 18 h. Under these two cases the reactor exhibited COD removal of 87 and 71% ensuring the effluent COD concentration of 520 and 1740 mg/L respectively. It reveals that,

when the reactor was loaded with COD of 6000 mg/L it experienced substrate inhibition and consequently biomass washout started. This state was attained obviously on account of high COD content in the influent wastewater. The values of COD removal percentage for various COD loading rates in the continuous study with synthetic wastewater are presented in Fig. 3. The said figure clearly reveals that COD removal percentage almost linearly decreases with the rise in COD loading rate.

The Food to Microorganism (F/M) ratio is a crucial factor affecting the performance of any biological reactor. In the EC-HUASB system the (F/M) ratio was calculated with respect to soluble COD and it expressed the mass of soluble COD available per unit biomass per day. The biomass generation was monitored by MLSS. In the initial condition the MLSS was 8,420 mg/L. Later, during the 17 stages of con-





Fig. 3. Plot of COD removal efficiency vs COD loading rate under continuous mode operation.

tinuous runs, MLSS continuously increased up to 21480 mg/ L. The easily digestible dextrose-based substrate caused the generation of this huge amount of biomass. To find out the influence of (F/M) ratio on COD removal, experimental values of those are plotted as shown in Fig. 4. It reveals that COD removal percentage almost linearly decreases with the rise in (F/M) ratio (COD basis). According to (51), 70% of the COD removal was attained at the loading rate of 3.5 kg COD/ m<sup>3</sup>/d. Similarly according to the (52), 92.6% of the COD removal was observed at the loading rate of 6.58 kg COD/m<sup>3</sup>/ d. In comparison to earlier studies, the EC-HUASB reactor raised the OLR by 50–80% for a desired COD removal efficiency in treating slaughterhouse wastewater.

## Biogas production:

Biogas production is one of the crucial parameters to estimate the performance of EC-HUASB reactor. To evalu-

ate performance of biogas production, the EC-HUASB reactor was operated under increasing OLR in respect of the varying HRTs. The HRT was gradually increased from 6 to 24 h to vary OLR between 2.40 and 9.60 kg COD/m<sup>3</sup>/d. During the study, an incremental biogas generation was obtained with appreciable methane gas percentage under various continuous runs. The biogas production was gradually increased in the overall operation ranging from 12.75 to 41.5 L/d with respect to the OLR 2.40 to 8.00 kg COD/m<sup>3</sup>/d, showing the CH<sub>4</sub> gas percentage ranging from 42 to 65%. In the first set of continuous runs, operated with the OLR 2.40 to 4.00 kg COD/m<sup>3</sup>/d, the biogas generation was enhanced from the 13.08 to 22.46 L/d, by virtue of reduction in HRT from 10 to 6 h.

Similarly, in the next set of continuous run the reactor was operated with the OLR between 4.00 and 6.00 kg COD/



Fig. 4. Plot of COD removal efficiency vs (F/M) ratio under continuous mode operation.

Table 2. Operational conditions for continuous operation in EC-HUASB reactor								
Run	Influent pH	Influent COD concentration	HRT	Steady state biomass concentration	OLR	(F/M) Ratio		
no.		(mg/L)	(h)	(mg/L)	(kg COD/m <sup>3</sup> /d)	(kg COD/kg MLSS/day)		
1.	7.02	1000	10	10400	2.40	0.23		
2.	7.04	1000	8	10750	3.00	0.28		
3.	7.06	1000	6	10950	4.00	0.36		
4.	7.04	2000	12	11300	4.00	0.35		
5.	7.03	2000	10	11700	4.80	0.41		
6.	7.06	2000	8	11900	6.00	0.50		
7.	7.05	3000	18	12500	4.00	0.32		
8.	7.07	3000	12	12750	6.00	0.47		
9.	7.06	3000	10	12900	7.20	0.56		
10.	7.05	4000	24	13150	4.00	0.30		
11.	7.04	4000	18	13600	5.32	0.39		
12.	7.07	4000	12	13950	8.00	0.57		
13.	7.06	4000	10	14100	9.60	0.68		
14.	7.08	5000	18	14350	6.65	0.46		
15.	7.07	5000	24	14750	5.00	0.34		
16.	7.08	6000	18	15300	8.00	0.52		
17.	7.07	6000	24	15500	6.00	0.39		

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m<sup>3</sup>/d that showed rise in the biogas generation from 24.02 to 38.01 L/d on account of reducing HRT from 12 to 8 h. In the third set of continuous runs with influent COD of 3000 mg/L, the biogas generation was observed to increase from 21.22 to 38.44 L/d due to variation in OLR from 4.00 to 7.20 kg COD/m<sup>3</sup>/d on account of HRT reduction from 18 to 10 h. In the fourth set of continuous runs with the influent COD 4000 mg/L, the OLR was in the range of (4.00–9.60) kg COD/m<sup>3</sup>/d, which caused biogas production between 20.68 and 47.65 L/d due to reduction in HRT from 24 to 10 h. In the next two

consecutive continuous runs the OLR was set between 5.00 and 8.00 kg  $COD/m^3/d$  due to HRT variation between (18–24) h. During this stage, the biogas production decreased from the level of 36.14 to 25.22 L/d due to the incremental COD concentration in the feed ratio. It is observed that with respect to the loading rate of 8.00 kg  $COD/m^3/d$  the maximum COD removal of 87% was attained along with the optimum biogas production for all the continuous runs with varying COD concentrations is presented in Fig. 5.



Fig. 5. Plot of biogas production rate vs COD loading rate under continuous mode operation.

The present study indicated that the EC-HUASB system was capable of treating the synthetic wastewater effectively. Before proceeding for slaughterhouse wastewater, the treatability of synthetic wastewater was undertaken to optimize the reactor efficiency with the freshly collected seed sludge. As Dextrose is a simple substrate in the synthetic carbonaceous wastewater the reactor was able to degrade the said wastewater very easily with substantial biogas production in absence of any inhibition. The maximum COD removal percentage of 96% was obtained at the loading rate of 4.0 kg COD/m<sup>3</sup>/d with respect to 24 h HRT. In later runs, when the OLR was increased to 5.32, 8.0 and 9.60 kg COD/m<sup>3</sup>/d further, the COD removal efficiency was reduced to 92, 87 and 78% respectively. Thereafter, in the last four continuous runs with OLRs 6.65, 5, 8 and 6 kg COD/m<sup>3</sup>/d, the COD removal percentage was marginally reduced to 74, 89, 84 and 76% due to the high amount of the influent COD concentration.

## Conclusion

The present study has been conducted to estimate the performance of EC-HUASB bioreactor for treating medium to high strength synthetic carbonaceous wastewater. Various experimental combinations were made to optimize COD removal efficiency under all the possible operating conditions. Experimental studies showed that mostly (69-96)% COD removal can be achieved in the EC-HUASB reactor under volumetric COD loading rate between (2.40-9.60) kg/  $m^{3}/d$  and (F/M) ratio (COD basis) of (0.23-0.68)  $d^{-1}$ . This is to note that appreciable COD removal is possible in the EC-HUASB system even under high volumetric COD loading rate, by virtue of additional attached biomass. It also reduced the (F/M) ratio (COD basis) and the amount of suspended biomass to be maintained in the EC-HUASB system. Under an OLR of 8.00 kg COD/m<sup>3</sup>/d the maximum COD removal percentage of 87% was observed with the optimum biogas generation of 41.52 L/d at the HRT of 12 h.

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