# Comparative Study on Start – Up Performance of HUASB and AF Reactors Treating Poultry Slaughterhouse Wastewater

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**ABSTRACT:** A study was performed in Hybrid Up flow Anaerobic Sludge Blanket (HUASB) and Anaerobic Filter (AF) reactors to compare the start-up time and optimum HRT required for the treatment of poultry slaughterhouse wastewater under similar loading conditions. Initially, the reactors were started at an OLR of 0.77 Kg COD/m<sup>3</sup>.d and HRT of 36 h. Loading rates were increased by reducing HRT 24, 16, 12, 10 and 8h which corresponds the OLR of 1.15, 1.74, 2.27, 2.74, 3.43 Kg COD/m<sup>3</sup>.d. HUASB reactor showed TCOD and SCOD removal efficiencies of 80% and 86%, respectively at an optimum HRT of 10 h whereas AF reactor showed 70% (TCOD) and 79% (SCOD) at optimum HRT of 12 h. Reducing HRT beyond 10h in HUASB reactor shown sludge wash out and lower COD removal efficiencies of less than 80% and beyond 12 h in AF marked decreased efficiencies as low as 66%. HUASB and AF reactors took 120, 147 respectively, for complete start-up. The granules of 2-2.5 mm sizes were observed in HUASB and less than 1 mm were observed in AF with settling velocities ranging between 0.5-0.83 m/min and 0.5 -0.65m/min, respectively. From Residence Time Distribution studies, dispersion numbers (<0.2) showed both the reactors attained plug flow regime. The present study revealed that the HUASB reactor has very good removal efficiency and less start-up time compared to that of AF reactor for the treatment of poultry slaughterhouse wastewater.

Keywords: Poultry Slaughterhouse wastewater, HUASB, AF, Granules, Settling velocity, Residence Time distribution

### INTRODUCTION

In India, rapid growth of poultry industry is leading to potential environmental pollution in terms of high Total Suspended Solids (TSS), Biochemical Oxygen Demand (BOD) and Chemical Oxygen Demand (COD), which is harmful if disposed off without treatment (Welch and Lindell, 1992). Basically, usage of water in poultry processing industry is high and the discharge may range from 5 to 10 gallons per bird with 7 gallons being a typical value (Kroyer, 1991). High rate reactors have been proven as very good options especially Anaerobic Filter (AF- Viraraghavan et al., 1990; Elmitwalli et al., 2000), Up flow Anaerobic Sludge Blanket (UASB- Lettinga et al., 1991; Leitao, 2004) and Hybrid UASB (HUASB- Kalyuzhnyi, 1997; Najafpour et al, 2006), for the treatment of poultry slaughterhouse wastewater (Masse et al.,

2000) since it contains sufficient nutrients for bacterial growth and recovery of methane during treatment. Anaerobic filters were used effectively for treating a variety of industrial wastewaters (Young, 1991). Treatment of slaughterhouse wastewaters has been reported by several authors using AF with random support (Henze and Hammeroes, 1983; Tritt, 1992; Del pozo et al., 2000) and UASB reactors (Sayed et al., 1984; Sayed et al., 1987; Manjunath et al., 2000; Torkian et al., 2003). In AF's, COD removal efficiencies varied between 80-90% for organic loads up to 20-25 kg COD  $/m^3$ .d. Ruiz et al., (1997) compared slaughterhouse wastewater treatment in UASB and AF, and found that the COD removal efficiency in UASB reactor was 90-60% at OLR up to 5 and 6.5 kg  $COD/m^3.d$ , respectively, whereas AF showed 63-84% efficiency at an

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OLR of 0.5-6 kg COD/m<sup>3</sup>.d. HUASB reactor is an anaerobic hybrid reactor offers the advantages of both UASB and AF's. This type of reactor is more suitable for the treatment of a series of soluble or partially soluble wastewaters (Tilche and Vieira, 1991). Several studies have been carried out on the start-up and performance of hybrid reactors (Shivayogimath and Ramanujam, 1999; Coates *et al.*, 1990). Treatment of hog slaughterhouse wastewater in hybrid reactors (polyurethane foam- packing media) (Borja *et al.*, 1993) showed COD removal efficiency of 90.2 and 93.4% at an OLR between 2.49 and 20.82 Kg COD/m<sup>3</sup>.d at HRT of 0.5 days.

Start-up is considered as difficult phase in anaerobic digestion because finding of optimum HRT is highly significant, to avoid the viable biomass washout. Hutnan et al., (1999) compared start-up and performance of UASB, HUASB and Anaerobic Baffled Reactor (ABR) and found that significant biomass washout was observed only in UASB and HUASB reactors at an OLR of 6 and 12 kg COD/m<sup>3</sup>.d respectively. Di Berardino et al., (1997) studied the start-up of Hybrid Filter using food processing wastewater at 25°C and 30°C and observed that the COD removal efficiencies were 60 and 83% respectively. Biogas generation was 30-170% higher at 30°C than 25°C. Residence Time Distribution (RTD) studies are used to evaluate the flow characteristics inside the reactor (Levenspiel, 1991; Show and Tay, 1999). In anaerobic reactor, effective substrate conversion is more important and it is optimum at intermediate degree of mixing (Smith et al., 1996). Though several studies have been carried on different types of slaughterhouse wastewater (hog, meat, cattle, sheep, goats) using different reactor configurations, studies on start-up strategy and granules formation using HUASB and AF reactors are scant. Hence, the present work was carried out with two main objectives; 1) to study the startup time and optimum HRT required for two different types of high-rate anaerobic reactors (HUASB and AF) working under similar conditions with poultry slaughterhouse wastewater and 2) to study the biomass properties like granulation period, settling velocities, specific gravity, SVI and SMA of sludge. Finally, RTD studies were conducted to study the type of flow inside the reactor at the end of the start-up.

## **MATERIALS & METHODS**

Bench scale continuous HUASB and AF reactors were made of Poly Vinyl Chloride (PVC) were used in the present study. Each reactor had an internal diameter of 10 cm and total height of 82 cm resulting in total volume of 6.4 L and working volume of 5.4 L with a gas head space of 1 L. The configuration of both the reactors was same except the packing media. For HUASB reactor, top one third of the reactor was filled with the media whereas the AF was packed with 4 L of packing media. The packing media was pleated PVC rings. The reactors were fed with substrate using peristaltic pump (Model: PP-20, Miclins). The peristaltic pump can maintain constant flow rate in the range of 2 mL/h to 10 L/h, available with timer and LED display for flow rate of function and time. The effluent pipe was connected to a water seal arrangement to prevent escape of the gas through the effluent. The configuration and the system layout are depicted in (Fig. 1). Five sampling ports were installed along the length of each reactor at 11 cm intervals, starting from a height of 5 cm above the reactor bottom. Biogas produced from the reactors was collected by water displacement method using Mariotte bottle. The operating temperature of the reactors was in the mesophilic range (29 -35°C).Poultry Slaughterhouse wastewater generated from Supreme Sugana Food Company located at Udumalpet, Tamilnadu, India was used as substrate. The wastewater was collected after a fat separator to avoid the hindrance of fat in the anaerobic digestion process. The wastewater used as feed was maintained in a refrigerator at 4°C. It was maintained in a feed reservoir and mixing was performed manually at regular interval. The substrate used for the experiment was combined slaughterhouse process wastewater. Characteristics of the wastewater are summarized in (Table1). The reactors were seeded anaerobically with a non granular sludge obtained wastewater treatment plant of the poultry slaughterhouse industry. The characteristics of seed sludge were analyzed as per standard methods (APHA, 1992). The Total Solids (TS), Total Suspended Solids (TSS), Volatile Suspended Solids (VSS), and ratio of VSS/ TSS are found to be 28,000, 14400, 10200 mg/L and 0.55 respectively.



Fig. 1. Schematic arrangements of HUASB and AF reactors

Table1. Characteristics of poultry slaughter house wastewater

Sl.	Characteristics	Values (mg/L)	
No.			
1	pH	7 - 7.6	
2	Colour	Brownish grey	
3	Total Solids (mg/L)	1400 - 3900	
4	Total Suspended Solids (mg/L)	300 - 950	
5	Total Volatile Solids (mg/L)	800 -1800	
6	$BOD_5(mg/L)$	750 - 1890	
7	Total COD (mg/L)	3000 -4800	
8	Soluble COD (mg/L)	1030 -3000	
9	VFA as acetate (mg/L)	250 - 540	
10	Alkalinity (mg/L)	600 - 1340	
11	Phosphates (mg/L)	16 - 32	
12	TKN (mg/L)	109 - 325	
13	Oil and Grease (mg/L)	800 - 1385	
14	Protein (mg/L)	580 -1000	

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(TSS), Volatile Suspended Solids (VSS), and ratio of VSS/TSS are found to be 28,000, 14400, 10200 mg/L and 0.55 respectively. Pleated PVC ring was selected as packing media because of its pleated surface can retain more biomass on surfaces rather than plain surfaces. The packing media had the dimensions of length, internal and external diameter 1.5, 1.1 and 1.3 cm respectively; porosity 98% and surface area 267 m<sup>2</sup>/m<sup>3</sup>. Both HUASB and AF reactors were started with an OLR of 0.77 Kg COD/m<sup>3</sup>.d. Throughout the start-up period feed Total COD (TCOD) was maintained, approximately 1150 mg/L after diluting with tap water. The initial HRT was 36h for both reactors. The loading pattern was increased stepwise by reducing the HRT at 24, 16, 12, 10 and 8 h. At each operating OLR, the reactors were operated continuously so as to reach pseudo steady state conditions, i.e. effluent COD and methane production remained relatively constant. Seed sludge comprised of 10200 mg/L of VSS with a low sludge loading rate of 0.11 Kg COD/Kg VSS.day. The specific Methanogenic Activity (SMA) of sludge was 0.09 g CH<sub>4</sub> COD/g VSS.day. Total and Soluble COD (for the soluble COD, a sample was filtered with a gas micro filter 0.45 um size), Volatile fatty Acids (VFA), Alkalinity, Volatile Suspended Solids (VSS), Volatile Solids (VS), Total Suspended Solids (TSS), Total Kjeldhal Nitrogen (TKN), Ammonia Nitrogen, Phosphates, Oil and grease were analyzed as per procedure detailed in Standard methods (APHA, 1992). All experiments were performed in triplicate. COD of the samples were measured by dichromate reflux method. VFA were measured by distillation method. A 100 mL of filtered sample was distilled with 100 mL of distilled water and 5 mL of 1:1  $H_2SO_4$ . After the distillation 150 mL distillate was titrated with 0.1 N NaOH, using phenolphthalein as an indicator and the value is reported on the basis of acetic acid. The alkalinity of the samples was measured as CaCO<sub>3</sub> by titrating the samples with 0.02 N  $H_2SO_4$  at a pH of 4.5. TKN samples were taken in a Kjeldhal flask and 50 ml of reagent, consisting of sulphuric acid, potassium sulphate and copper sulphate, was added to it. The sample was digested completely, cooled and diluted with distilled water to 300 mL and then sodium hydroxide was added and distilled. The distillate was collected in boric acid and titrated against 0.02 N sulphuric acid using mixed indicator to get the end point. The ammonia-N was determined by direct Nesslerization method. The estimation of oil and grease was determined by partition gravimetric method.

The flow rate, pH and amount of biogas generated were recorded daily and other parameters TCOD, SCOD, VFA and Alkalinity were measured once in three days. The biogas composition was measured two times weekly by passing a known volume of biogas through a KOH scrubber. Protein was measured by multiplying the difference between TKN and NH<sub>4</sub>-N by 6.25 (AOAC, 1984). The Specific Methanogenic Activity (SMA) of the sludge was determined using the protocol as adopted by Isa et al., (1993) without addition of nutrients (Jawed and Tare, 1999). SMA was measured from the slope of the linear portion of cumulative methane production rate and the final VSS in the serum bottle. The SMA of sludge was measured for both initial seeding of the reactors and at the end of start-up. The VSS concentration along the reactor height and RTD studies were conducted at the end of start-up. 250 ppm Lithium chloride (LiCl) tracer was injected as a pulse input into the reactor at the end of start-up, i.e. at 120th day for HUASB and at 147th day for AF. Samples were collected every 4 h over a period of 48 h. The samples were analyzed (average of triplicate) using a flame photometer (Model: CL 354, Elico).

### **RESULTS & DISCUSSION**

During the initial 10 days, pH was varied between 6.6 and 7.0 in HUASB and 6.5 - 7.0 in AF reactor as shown in (Fig. 2 and 3). This was expected as the acid fermentation phase is always more rapid than that of methanogenic phase (Borja and Banks, 1995). However, after initial drop, consistent pH level of 7.2 - 7.8 were maintained in the effluent of both reactors indicating healthy environment. The variations of VFA and alkalinity are depicted in (Fig. 4 and 5). The alkalinities were increased in both the reactors when loading rates were increased. At the end of the startup phase, VFA was less than100 mg/L in both reactors R1 and R2 at an OLR of 2.74 and 2.27Kg COD/m<sup>3</sup>.d respectively. Throughout the start-up period, VFA/ Alkalinity ratio varied between 0.10-0.23 and 0.110.32 for HUASB and AF reactors respectively, except the initial few days. It had clearly shown that no instability occurred inside the reactors (Leitao, 2004).



Fig. 2. Variations of pH during start-up in HUASB reactor



Fig. 3. Variations of pH during start-up in AF reactor



Fig. 4. Variations of VFA and alkalinity during the start-up in HUASB reactor



Fig. 5. Variations of VFA and alkalinity during the start-up in AF reactor

The COD removal efficiencies of HUASB and AF reactors are shown in (Fig. 6 and 7). respectively. Initial applied OLR for both HUASB and AF reactor was 0.77 Kg COD/m<sup>3</sup>.day with an HRT of 36 h, to avoid the inoculated biomass washout (Hickey et al., 1991). The HUASB reactor was maintained for a period of 25 days in the same loading rate, after which the reactor has reached the steady state (variation in gas collection and removal efficiency is less than 3%) and removal efficiencies in terms of TCOD and SCOD was 25 and 30%, respectively. During the stepped increase of OLR, COD removal efficiencies were gradually increased. On day 98, the removal efficiencies of TCOD and SCOD were reached around 81 and 88% at a Total Organic Loading Rate ( $OLR_{T}$ ) of 2.74 and Soluble Organic Loading Rate (OLR<sub>s</sub>) of 1.18 Kg COD/ m<sup>3</sup>.d, respectively. This was in general agreement with COD removal efficiencies were increased with time of operation (Saravanane *et al.*, 2001). On day 111, Total COD removal efficiency was dropped less than 80% while further increasing  $OLR_{T}$  to 3.43 Kg COD/m<sup>3</sup>.d by reducing HRT to 8h. Reduction in COD removal efficiency at 8h HRT was attributed to sludge washout and subsequently HRT was increased to 10h and maintained for 9 days to reach steady state (i.e. up to 120th day). Finally, the HUASB reactor achieved the removal efficiencies of 80 and 86% in terms of TCOD and SCOD, respectively at the end of complete start-up. Maintaining 10 h HRT ensured that sufficient selective pressure for granulation and optimum contact time between

microbial and substrate available in the reactor (Lettinga *et al.*, 1997). In contrary to TCOD removal efficiency, the SCOD removal was 86 %, which is higher than TCOD. HUASB reactor took 120 days for complete start-up. The overall duration for complete start-up of HUASB reactor was comparable to the studies conducted by Ruiz et al. (1997) who achieved start-up time of 117 days while evaluating the treatment of slaughterhouse wastewater using UASB reactor.

After starting the AF with an OLR of 0.77 Kg COD/m<sup>3</sup>.d.ay, it was maintained in batch mode for 20 days and another 14 days it was continued in the same loading in continuous mode to enhance the growth of microbes on the media. As it was followed in HUASB reactor, the OLR's were increased stepwise to 1.15, 1.74, 2.27, 2.88, and  $3.43 \text{ Kg COD/m}^3$ .d. by reducing the HRT to 24, 16, 12, 10, 8h respectively. After 109 days, AF achieved the TCOD removal efficiencies of 72% at an OLR of 2.27 Kg COD/m<sup>3</sup>.d and 78 % in terms of SCOD at an OLR of 0.97 Kg COD/m<sup>3</sup>.d. Reduction in HRT less than 12h (i.e. 10 and 8h) showed decreased trend in COD removal efficiencies in terms of both TCOD and SCOD. This may be due to fact that during the short contact time between attached/suspended biomass and substrate; and ultimate reduction of entrapment capacity (Lettinga et al., 1997). Hence the optimum HRT was found to be 12 h for AF reactor and it took 147 days for complete startup. At the end of start-up removal efficiencies of TCOD and SCOD were 70% and 79%, respectively. The prolonged start-up may be attributed, due to low SMA of the sludge used for seeding of the reactors as well as it was needed much time to form the microbial growth onto the filter material. The biogas production with respect to loading rates was shown in (Fig. 8 and 9). It can be seen from graph, the gas production was progressively increased with increasing OLR. The maximum gas collection of 3.3 and 2.4 L/d were observed in HUASB and AF reactors at an OLR of 2.76 and 2.27 Kg COD/m<sup>3</sup>.d and the methane content was around 75 and 56%, respectively. The SMA of seed sludge used in both reactors was 0.09 g CH<sub>4</sub>-COD/g VSS.d. At the end of startup, the SMA values were found to be 0.17 and 0.14 g CH<sub>4</sub>-COD/g.VSS.d in HUASB and AF reactors, respectively. The SMA values of HUASB could not be compared with AF since the sludge taken from AF was only at the suspended portion and not from the attached portion. The suspended portion has only given partial results. Though the lower methanisation of suspended sludge observed in AF compared to HUASB reactor, the removal efficiencies observed were quite good in the range of 39 to 70%.

HUASB reactor showed better removal efficiencies of TSS of 60-84% up to an OLR of 2.88 Kg COD/m<sup>3</sup>.d. But in contradictory to HUASB reactor, AF showed the removal efficiencies as low as 37-71% up to an OLR of 2.27 Kg COD/m<sup>3</sup>.d. Reduction in HRT from 10 to



Fig. 6. Total COD and soluble COD removal during the start-up of HUASB reactor



Fig. 7. Total COD and Soluble COD removal during the start-up of AF reactor



Fig. 8. Organic loading pattern and biogas production during start-up of HUASB reactor



8 h showed marked decrease of TSS removal efficiency from 84 to 73% in HUASB reactor. But reduction in HRT from 12 to 10 h showed TSS removal efficiency from 71 to 62% in AF reactor. Further reduction of 8 h HRT led to lower removal efficiencies as low as 62% and 51% in AF reactor. It was clearly shown that the hydrolysis and entrapment of TSS were more in granular sludge compared to media bio-film of AF reactor. Moreover, the action of top filter media of HUASB reactor prevented the washout of solids. The VSS profiles of HUASB and AF reactors at the end of start-up were shown in the (Fig. 10). It can be seen from the figure that higher VSS concentration of 27000 mg/L and 3200 mg/L were in the lower (14 cm height from bottom) and 3000 mg/L and 2000 mg/L were in the upper part (58 cm height from bottom) of the HUASB and AF reactors, respectively. Majority of solids attributed in lower part of the reactors, which is in correlation with findings of treatment of slaughterhouse wastewater using UASB reactor (Torkian *et al.*, 2003). In the AF, it was unable to determine the VSS of attached biomass, however the VSS of suspended growth were determined and it was observed as 12% and 66% at bottom and top portions of HUASB reactor.



Fig. 10. Profile of VSS at various heights at the end of start-up in HUASB and AF reactors

The depth of sludge bed in HUASB was ascertained by drawing the sample from sample port since the reactors were made up of PVC material. Initially the sludge bed depth was observed above 2<sup>nd</sup> port from bottom. The height of sludge bed got reduced to below 2<sup>nd</sup> port at an OLR of 1.15 Kg COD/m<sup>3</sup>.d, because of flocculant sludge washout. At an OLR of 2.27 Kg COD/ m<sup>3</sup>.d height of sludge bed remained below 2<sup>nd</sup> port (< 25 cm). After reaching the OLR of 2.74 Kg COD/m<sup>3</sup>.d, it was further increased to 3.43 g COD/ 1.d by decreasing HRT to 8 h and granulation were observed with sludge wash out. Increase in the sludge formation increased the height of sludge bed above 2<sup>nd</sup> port i.e. 25 cm at the end of startup period. Similar observation of increasing OLR led to increase the sludge bed height was noticed during the treatment of palm oil mill effluent in HUASB reactor (Najafpour et al., 2006). Finally, by day 120, concentrated granular sludge with good settleability was formed in HUASB reactor. In AF reactor sludge depth had not been considered since the type of biomass observed was attached growth system.Scmidt et al. (1996) have reported the settling velocities as poor, satisfactorily and good settling fraction with settling velocities ranges of 20m/h, 20 to 50 m/h and above 50 m/h, respectively. The granular sludge obtained from HUASB and AF reactors were shown in (Fig.11a and 11b). Settling velocities of sludge granules varied between 0.5 to 0.83 m/min for granule sizes 2 to 2.5 mm in HUASB reactor, and found to be satisfactory. However, in the AF reactor, settling velocities of granules varied from 0.5 to 0.65 m/min which showed very low settling velocities. Escaping of suspended sludge can be prevented through the media due to the action of filtration mechanisms. The size of suspended granules found in AF was around 1 mm.

The specific gravity of sludge taken from HUASB and AF reactors were 0.8 and 0.74 respectively and the results are comparatively lower than the treatment of slaughterhouse wastewater using UASB reactor (i.e., 1.02 to 1.14) as reported by Torkian *et al.*, (2001). In both



Fig. 11a. Granules obtained from HUASB reactor



Fig. 11b. Granules obtained from AF reactor

Table 2. Dispersion number at the end of startup in the reactors						
Reactor	OLR (Kg OD/m <sup>3</sup> .d)	Influent velocity (m/d)	Hydraulic Residence Time (h)	Dispersion Number(D/µL)	Peclet Number (reciprocal of	
					Dispersion number)	
HUASB	2.74	1.65	10	0.19	5.26	
AF	2.27	1.38	12	0.15	6.67	

Table 2. Dispersion number at the end of startup in the reactors

HUASB and AF reactors initial SVI was 35 mL/g which was further reduced to 20 mL/g in HUASB reactor and increased to 45 mL/g in AF reactor, at the end of start-up period. These results are comparable with that the performance of UASB reactor treating distillery spent wash and reported as SVI of 20-40 mL/g (Sunilkumar, 1988). To identify the combined effect of liquid velocity, gas production and presence of sludge mixing patterns, the tracer studies were carried out at the end of start-up period an up flow velocity of 1.65 and 1.38 m/d for HUASB and AF reactors, respectively. The normalized concentration against normalized time is shown in (Fig.12). The calculated dispersion numbers are shown in (Table 2). It was observed from the dispersion number that the  $D/\mu L$  values were less than 0.2 indicated the flow pattern is plug flow in both reactors (Levenspiel, 1991). This could be due to low gas production and less liquid up flow velocities.



Fig.12. Residence time distribution curve at the end of start-up in HUASB and AF reactors

### CONCLUSION

This study reveals that under the same substrate and similar loading conditions, HUASB reactor performed well compared than AF reactor, during the entire start-up period. The TCOD and SCOD removal efficiencies were as high as 80% and 86% in HUASB as compared with AF of 70% and 78%, respectively. The optimum HRT was found to be 10 and 12h, at loading rates of 2.74 and 2.27 Kg COD/m<sup>3</sup>.d for HUASB and AF reactors respectively. The granulation (2 to 2.5 mm) and top portion of attached microbial populations of HUASB reactor had contributed the higher TCOD and SCOD removal efficiencies. The settling velocities of sludge obtained from HUASB and AF reactors were ranging between 0.5-0.83 m/min and 0.5 -0.65m/min, respectively. The SMA of AF was less than that of sludge obtained from HUASB reactor indicated low methanogenic activity was prevailing in AF reactor. The Residence Time Distribution study revealed that the type of flow was plug flow regime due to low biogas production as well as lower up flow velocities. To conclude, HUASB reactor was needed less time for start-up and showed better removal efficiencies as compared to AF reactor using the same substrate of poultry slaughterhouse wastewater.

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