PERFORMANCE EVALUATION OF FULL SCALE UASB REACTOR IN TREATING SEWAGE UNDER PSYCHROPHILIC CONDITIONS

A Thesis

Submitted in partial fulfillment of the requirements for the award of the degree of

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IN

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With specialization in

ENVIRONMENTAL ENGINEERING

Under the supervision of

Prof. Veeresh Gali

By

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to

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CERTIFICATE

This is to certify that the work which is being presented in the project title **PERFORMANCE EVALUATION OF FULL SCALE UASB REACTOR IN TREATING SEWAGE UNDER PSYCHROPHILIC CONDITIONS** in partial fulfillment of the requirements for the award of the degree of Master of technology in civil engineering with specialization in **Environmental Engineering** and submitted in Civil Engineering Department, Jaypee University of Information Technology, Waknaghat is an authentic record of work carried out by **Manisha Thakur (142753)** during a period from August 2015 to July 2016 under the supervision of **Dr. Veeresh Gali,** Professor, Civil Engineering Department, Jaypee University of Information Technology, Waknaghat.

The above statement made is correct to the best of my knowledge.

Date: 06-06-2016

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ABSTRACT

The aim of the study was to assess the performance of a full scale Upflow Anaerobic Sludge Blanket reactor (UASBR) with respect to COD removal for domestic wastewater under low temperature conditions. Tests on Temperature, pH, Alkalinity, BOD, COD, Total Solids (TS) and Volatile Solids (VS) were conducted for influent and effluent to evaluate the performance of reactor under psychrophilic conditions. Grab samples were collected once a month from sewage treatment plant (STP) located at Lalpani, Shimla, and Himachal Pradesh, India .

The COD removal efficiency ranged from 13 - 42%, which was not satisfactory. The maximum removal efficiencies of BOD, Suspended Solids (SS) and VS were 36%, 82% and 67% respectively. The main reason for the unsatisfactory performance of UASB reactor was the poor operation and maintenance of the reactor. Biochemical methane potential (BMP) of domestic wastewater was determined at different substrate to biomass (F/M) ratio in order to assess the biodegradability of wastewater. BMP test was performed in batch bioassay by using the serum bottle technique and gas was collected by inserting a glass syringe in the bottle. Granular sludge from UASB reactor was used as inoculum. Specific methanogenic activity (SMA) of reactor sludge was also determined at 20ºC to evaluate the quality of sludge. The SMA test was carried out by using serum bottle technique using sodium acetate as substrate. A COD mass balance was done for BMP.

BMP of substrate was influenced by F/M ratio. A decreasing trend was noticed in the BMP values when F/M ratio decreased from 4 to 0.35 at both controlled and uncontrolled conditions. BMP at constant temperature (19^{o}C) was found to be more than BMP at ambient temperature (0 – 26^{o}C) because the sudden increase (i.e., shock) in temperature leads to decrease in bio-methane producing bacteria. F/M ratio 2 was found optimal. The methane producing capability of sludge at F/M ratio 2 is more as compared to F/M ratios 1 and 0.5.

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Manisha Thakur

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NOTATIONS AND ABBREVIATIONS

AF	Anaerobic Filter
APHA	American Public Health Association
ASP	Activated Sludge Process
AWWA	American Water Works Association
AWWT	Anaerobic Wastewater Treatment
BMP	Biochemical Methane Potential
BOD_5	Biological Oxygen Demand for 5 Days @ 20ºc
CH_4	Methane
CO_2	Carbon Dioxide
COD	Chemical Oxygen Demand
COD_{col}	Colloidal Chemical Oxygen Demand
COD_{dis}	Dissolved Chemical Oxygen Demand
COD_{ss}	Suspended Chemical Oxygen Demand
COD_t	Total Chemical Oxygen Demand
CPCB	Central Pollution Control Board
CPHEEO	Central Public Health Environmental Engineering Organisation
DO	Dissolved Oxygen
F/M	Food and Microorganisms ratio
GOI	Government of India
HPPCB	Himachal Pradesh Pollution Control Board
HRT	Hydraulic Retention Time
I&PH	Irrigation and Public Health
MoEF	Ministry of Environmental Forest
N	Nitrogen
NH_3	Ammonia
O_2	Oxygen

OP	Oxidation Pond
P	Phosphorous
SBR	Sequencing Batch Reactor
SMA	Specific Methanogenic Activity
SRT	Solid Retention Time
SS	Suspended Solids
STP	Sewage Treatment Plant
STP	Sewage Treatment Plant
TS	Total Solids
UASB	Up flow Anaerobic Sludge Blanket
VSS	Volatile Suspended Solids
WEF	Water Environment Federation
YAP	Yamuna Action Plan

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CHAPTER 1

INTRODUCTION

Anaerobic treatment technology has been widely used in developing countries for last two decades. Introduction of unconventional bioreactors i.e., anaerobic fixed bed, anaerobic fluidized bed and Upflow Anaerobic Sludge Blanket reactors has contributed to great success in the development of anaerobic wastewater technology (Uemura et al., 2000). Even though the performance of anaerobic technology in treatment of different types of industrial wastewater is satisfactory, but is still facing challenges in its applicability to lower strength wastewaters i.e., sewage and domestic wastewater (Van Der Last and Lettinga, 1992; Uemura et al., 2000).There are different treatment technologies which are adopted for the treatment of sewage in India, which includes Activated Sludge Process (ASP), Upflow Anaerobic Sludge Blanket (UASB), Oxidation Pond (OP) and advanced technologies like Sequencing batch reactor (SBR), Membrane bioreactor (MBR). Anaerobic sewage treatment is more suitable at mesophilic temperature conditions (>20ºC) because of high production of energy in the form of methane. However, treatment of sewage at psychrophilic temperature conditions (<20º) is facing challenges.

UASB reactor is an anaerobic biological reactor used to treat all types of high-strength wastewater (e.g. agro-industrial waste, pharmaceutical waste, textile waste, etc.) or as decentralized treatment systems for domestic wastewaters (e.g. domestic sewage). It was developed by Lettinga in Netherlands in 1970. Since then UASB reactor has been studied worldwide in a number of pilot and full scale systems. The UASB technology has been extensively employed for low strength wastewater treatment in India. In India 47 UASB based STPs are in operation and about 4 UASB based STPs are under construction and commissioning phase (CPCB, MoEF, 2015). The Government of India initiated the Yamuna Action Plan (YAP) in 1993 for conservation of river Yamuna under which 16 UASB STPs were commissioned (Khalil et al., 2008; Sato et al., 2006). Location of STPs in YAP with treatment capacities is shown in Figure 1.1. Earlier, UASB plants were provided with post-treatment unit, i.e., polishing ponds, but now new options are being surveyed to meet the stringent regulations (Khalil et al., 2008).

Previous studies on the performance of UASB reactors had revealed that at low temperatures (5-20ºC), hydrolysis of entrapped solids which agglomerate in the sludge bed when high loading rates are applied, is the reason which limit the process (Zeeman et al., 1999).

Accumulation of solids will require a regular sludge discharge. That being so, there will be increase in excess sludge retention time (SRT) and concurrently less stabilized sludge bed with a low specific methanogenic activity (SMA) which will result in poor soluble COD removal (Mahmoud 2008).

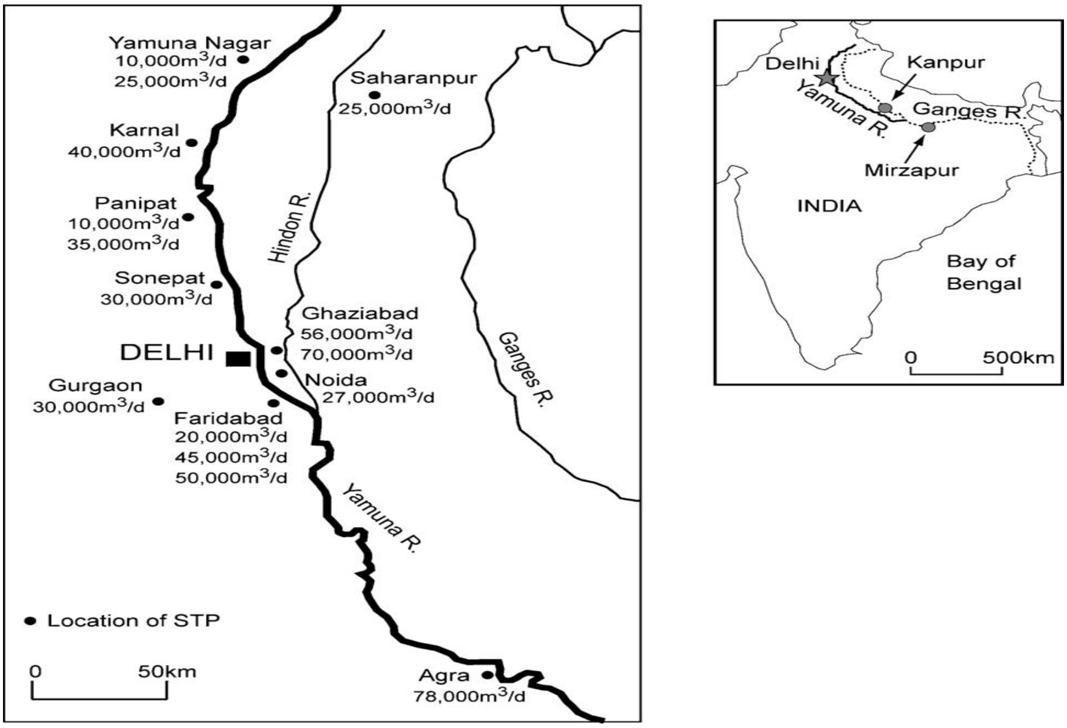

Figure 1.1: Location of STPs with Treatment Capacities [Source: Sato et al., 2006]

Biochemical methane potential (BMP) is one of the most common parameters for assessing the biodegradability of waste. It has become an interesting tool for waste characterization to determine the potential of its methane production (1 methane/g COD removed) (Angelidaki et al., 2009; Kaosal et al., 2012). BMP tests are widely applied to figure out the information about the methanogenesis of specific substrates and provide results which are necessary to calibrate and validate mathematical models. It is a simple and cost effective procedure which is carried out under anaerobic conditions in bench scale to determine the amount of biogas produced per gram of volatile solids (VS) present in substrates (Hussain et al., 2015).

$$BMP = \frac{Maximum\ Cumulative\ Methane\ (mL)}{g\ COD\ removed}$$

Specific methanogenic activity (SMA) evaluates the methane producing capability of sludge by using specific substrate (Hussain et al., 2013). SMA also determines the methanogenic and relative microorganism's levels for anaerobic sludge under different operating conditions (Javed et al., 1999). It is performed under anaerobic conditions in bench scale and in batch process. The SMA is obtained by representing methane production (in g COD) against time and divided by g VSS added (Hussain et al., 2015). The experimental set up for BMP and SMA test is shown in Figure 1.2.

$$SMA = \frac{Methane\ produced\ (gCH4 - COD)}{g\ VSS\ (added)}$$

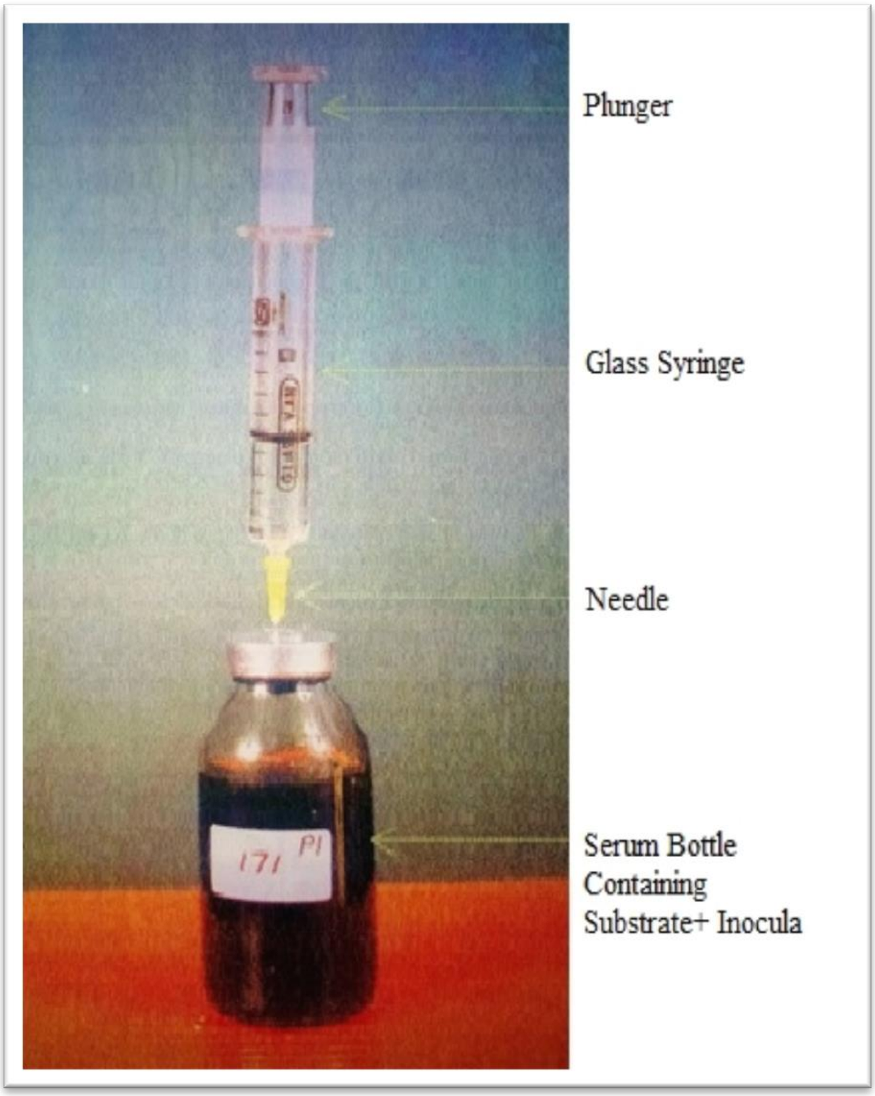

Figure 1.2: BMP and SMA Experimental Set up

1.1 STUDY AREA

1.1.1 General

Shimla is the capital city of Himachal Pradesh. It lies on a ridge and is spread on its seven spurs. The city length is app. 9.2 km. The city is an 18 sq. km mountainous region whose elevation from the sea level is 7866.10 ft. or 2397.59 meters. It lies in the south-western ranges of the Himalayas at 31.61ºN 77.10ºE. The top of Jakhoo Hill is the highest point of Shimla which is about 8051 ft. or 2454 meters. The nearest water body is the Sutlej River which is 21 km away from the city. Map of Shimla district is shown in Plate 1.1.

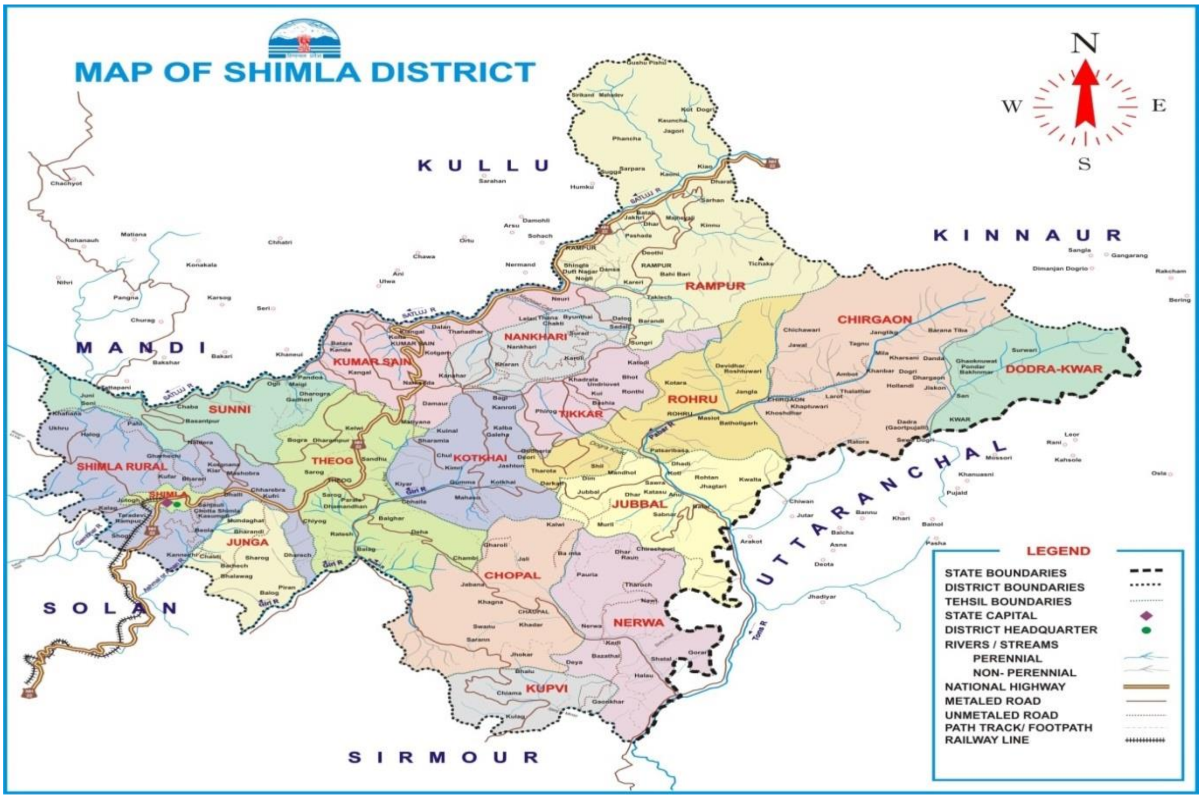

Plate 1.1: Map of Shimla district [Source: http://hpshimla.nic.in/sml_hist.htm]

1.1.2 Climate

The winter temperature in Shimla varies from 18ºC to - 4ºC and in summer from 32ºC to 6ºC. The city receives the monsoon during the months of July to September with annual average rainfall of about 150mm.

1.1.3 Demographics

As per provisional data of 2011 census Shimla urban agglomeration had a population of 171,817.

1.2 SEWERAGE SYSTEM

Shimla Municipal area has a well-planned underground sewerage system and is properly maintained by I&PH department. The first sewerage network in the Shimla city was laid in the year 1880 to serve 18000 populations. In year 2005, under the assistance from OPEC and state funding, a new sewerage system network was designed and implemented to cater the demand for 2031 year in Shimla.

1.2.1 Sewage Treatment Plants

I&PH have constructed 6 STPs with total capacity of 35.63 MLD through OPEC funding. I&PH have given operation and maintenance of these STPs on management contract. The details of treatment plants are presented in Table 1.1.

Table 1.1: Details of Sewage Treatment Plants

S.No	Name Of Sewerage Treatment Plant	Capacity (MLD)	Technology Used
1	Lalpani	19.35	UASB
2	Sanjauli Malyana	4.44	Extended Aeration System
3	Dhalli	0.76	
4	Snowdown	1.35	
5	North Disposal	5.80	
6	Summer Hill	3.93	

(Source: IPH report on 19.35 MLD STP at Lalpani)

1.2.2 Lalpani Sewage Treatment Plant

1.2.2.1 Location of STP

Lalpani STP is located near Baragaon on Shogi bypass as shown in Plate 1.2.

Plate 1.2: Location of Lalpani Sewage Treatment Plant

1.2.2.2 Lalpani Sewerage Zones, Population and Sewage Quantity

Lalpani sewerage zones cover different areas of Shimla town having different residential population. The contributory populations from various Water Supply zones are given in Table 1.2.

Table 1.2: Lalpani Sewerage Zones, Population and Sewage Quantity

Sr.	Water Supply Zones	Density wise residential population					
		High	**Medium**	**Low**	**Complex**	**Govt. Est.**	**Total**
1.	Kasumpati	13,510	3,515	1,780	126	-	18,931
2.	Mansfield	15,498	912	-	-	-	16,410
3.	B.C.S	17,535	5,850	5,788	-	-	29,173
4.	Sanjauli	24,320	372	-	-	-	24,692
5.	Ridge	4,632	23,160	-	3,010	7,720	38,522
6.	High Court	-	36,253	-	-	-	36,253
7.	Chakkar	-	4,095	439	2,573	-	7,107
8.	Kamna Devi	-	984	1,200	-	-	2,184
9.	Viceregal Lodge	14,335	2,458	18,400	-	-	35,193
10.	A.G. Office	17,072	6,789	11,733	-	-	35,603
	Total	1,06,902	84,397	39,340	5,709	7,720	2,44,068

(Source: IPH report on 19.35 MLD STP at Lalpani)

1.2.2.3 Flow Calculations

The wastewater generated is taken as 80% of the water supplied in the contributory area. The wastewater generated in the year 2031 & 2016 on the basis of the contributory population given in Table 1.2 is tabulated in Table 1.3. Population for year 2016 is 2/3 that of the year 2013 and wastewater generated in 2016 is 19.35 MLD.

Table 1.3: Wastewater generation in 2031 and 2016

Sr.	Population	Population in 2031	Total water Demand in 2031	Wastewater generated	
	Type	(nos.)	(nos.)	2031 (MLD)	2016 (MLD)
1	Permanent	1,77,793	26.669	21.335	14.22
2	Floating	66,275	6.627	5.302	3.53
3	Others		3.361	2.689	1.793
	Total	2,44,068	36.657	29.02	19.35

(Source: IPH report on 19.35 MLD STP at Lalpani)

1.3 TREATMENT PROCESS

The treatment process at the Lalpani STP consists of different treatment unit i.e. Inlet Chamber (Plate 1.3), Screen Chamber (Plate 1.4), Grit Chamber (Plate 1.5), UASB Reactor (Plate 1.6), Extended Aeration Tank (Plate 1.7), Secondary Clarifier (Plate 1.8), Flash Mixer, Clariflocculator, Sludge Pumping Stations, Distribution Boxes, and Filter Press.

The inlet chamber receive raw sewage and pass it further to screen channel and grit channel where the floating matters are trapped and removed in screen channel and grit is removed in grit channel. This treatment is known as primary treatment. After primary treatment the screened sewage is treated biologically in UASB reactor followed by extended aeration process comprising of aeration tank and secondary settling tank.

During winter season due to fall in temperature the removal efficiency of extended aeration process is reduced and effluent from secondary settling tank is treated physico-chemically by adding alum in flash mixer and settling out the flocs in Clariflocculator. The sludge from UASB reactor and secondary settling tank is then dewatered using filter press for further disposal.

Plate 1.3: Inlet Channel

Plate 1.4: Screen Channel

Plate 1.5: Grit Channel

Plate 1.6: UASB Reactor

Plate 1.7: Aeration Tank

Plate 1.8: Secondary Clarifier

1.4 ANAEROBIC WASTEWATER TREATMENT

Anaerobic process is a biological process carried out in the absence of oxygen for the stabilization of organic matter by conversion to methane and inorganic products such as carbon dioxide and ammonia.

$$\textit{Organic materials} + \textit{Nutrients} \longrightarrow CH_4 + CO_2 + NH_3 + \textit{Biomass}$$

Anaerobic wastewater treatment (AWWT) such as the Anaerobic Filter (AF) and the Upflow Anaerobic Sludge Blanket (UASB) offers number of advantages in comparison with conventional aerobic treatment for wastewater. Benefits and drawbacks of AWWT are listed in Table 1.4.

Table 1.4: Benefits and drawbacks of anaerobic wastewater treatment over conventional aerobic methods

Benefits	1.	Excess Sludge (stabilized) production is low.
	2.	Energy is not required for aeration.
	3.	Low nutrient requirements.
	4.	Production of biogass (methane).
	5.	Compounds like ammonia are conserved, which in specific cases might represent an important benefit.
	6.	Process can handle high space loads.
Drawbacks	1.	Anaerobic bacteria are very susceptible to inhibition by large number of compounds.
	2.	Slow start-up process if adapted seed sludge is not available.
	3.	Anaerobic treatment depends on adequate post- treatment for proper removal of BOD, ammonia and nutrients.

(Source: Lettinga et al., 1984)

1.4.1 Anaerobic Microbial Degradation

In anaerobic degradation four metabolic groups of bacteria are distinguished which are:

(1) Hydrolytic bacteria which resolve polymers such as proteins and carbohydrates into monomers; (2) Fermentative bacteria which help in fermentation of these monomers into alcohols, acids, carbon dioxide, hydrogen and ammonia; (3) Acetogenic bacteria which convert higher volatile fatty acid and alcohols into acetic acid and hydrogen, and (4) Methanogenic bacteria which utilize methanol, acetate, hydrogen and carbon dioxide to produce methane (Lettinga et al., 1984). A schematic of reaction steps is outlined in Figure 1.3.

1.4.2 UASB Reactor

Lettinga develop Upflow Anaerobic Sludge Blanket Reactor (UASB) in 1970s in the Netherlands. UASB is a suspended growth system and require a proper hydraulic and organic loading rate in order to facilitate the granulation which is also known as dense biomass aggregation. The granules are bigger in size (1-3 mm diameter) and heavier, which made them to settle down and to retain within the reactor. The biomass concentration in the reactor may become as high as 50 g/L. Thus, even at a very low HRT of 4 hours very high Sludge Retention Time (SRT) can be achieved. At top of reactor three phase separation between gas-liquid-solid takes place. Any biomass leaving the reaction zone is directly recirculates from settling zone. Figure 1.4 and Plate 1.9 shows a typical cross section of UASB reactor and full scale UASB reactor located in Lalpani STP.

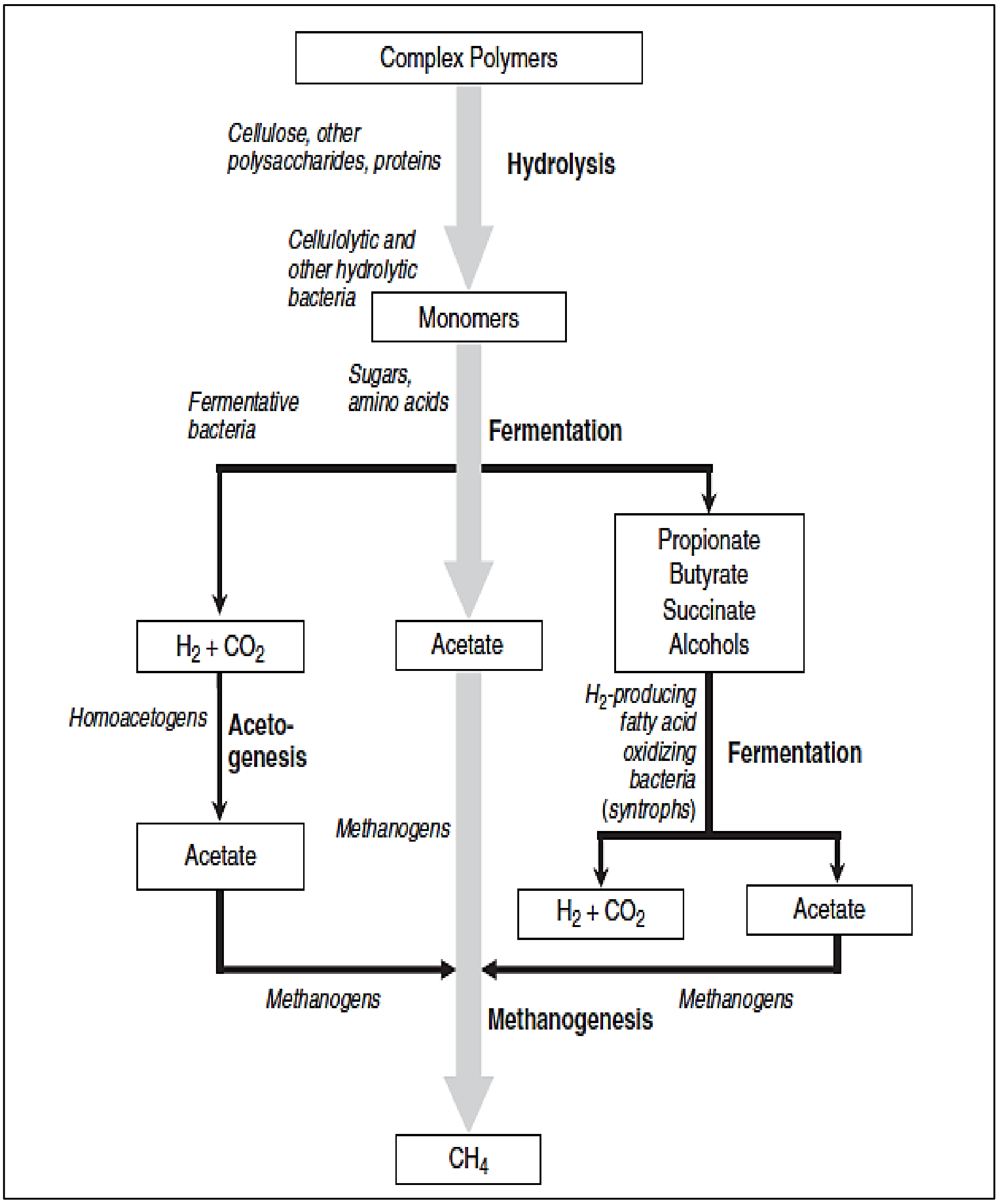

Figure 1.3: Anaerobic Digestion (Source: Biogas Technology for sustainable Second Generation Biofuel Production, 15-19 August 2011)

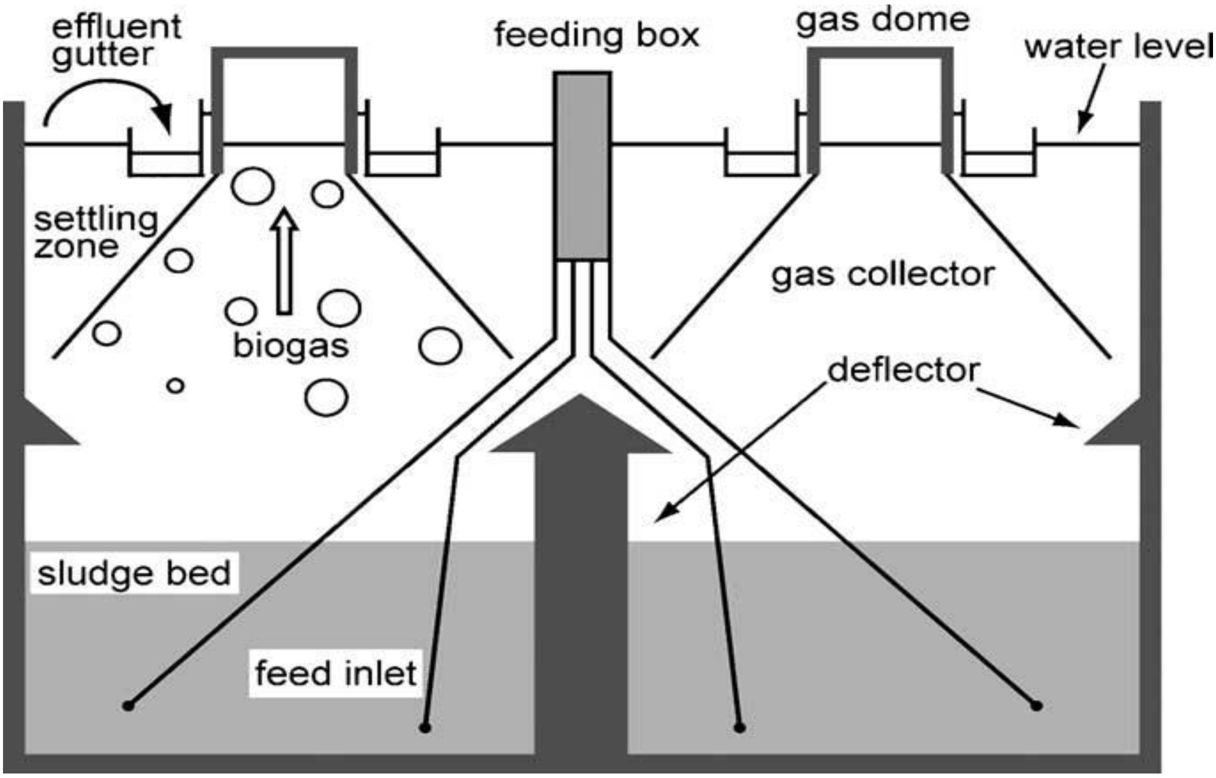

Figure 1.4: Sectional View of UASB Reactor [Sato et al., (2006)]

Plate 1.9: Full Scale UASBR located in Lalpani STP

CHAPTER 2

LITERATURE REVIEW

Great success in development of anaerobic wastewater treatment has provided various methods for the treatment of sewage. One of the most extensively and successfully used anaerobic system is UASB. Its suitability to lower-strength wastewater at low temperature is a big upcoming challenge for anaerobic treatment. Therefore, several studies have been conducted for low-strength wastewater at low temperature in both pilot plant and full scale plant.

2.1 STUDIES ON UASB PILOT PLANT

Singh et al., (1998) analysed the probability involved in starting and operating UASB reactors with a municipal wastewater at a lower temperature of 20ºC. Study concluded that the UASB system could be a technically feasible alternative in the treatment of municipal wastewater in mild temperature region. A start-up period of more than 60 days would be required to achieve a steady condition (COD removal efficiency of about 80 to 85%). The feasibility of sewage treatment by an UASB reactor was also studied by Uemura et al., (2000) using actual sewage at a fixed HRT of 4.7h and at temperature in the range of 25-13ºC for six months. The result showed a satisfactory performance, achieving a total COD removal of 69.4% based on total influent and soluble effluent, on, average throughout the entire experiment. Above two studies have been done with inoculum sludge whereas Kalogo et al., (2001) investigate the dynamics of a self-inoculated UASB reactor treating sewage and results showed that after 22 weeks of operation at 29ºC with an HRT of 4h, the reactor removed total COD, soluble COD and SS up to 80%, 60%, and 90%. The results confirm that without inoculation, the operation of a UASB reactor on raw domestic sewage is feasible, yet with slow biological conversion. When high loaded reactors were applied, the produced sludge was not stabilised and needs further stabilization in a separate digester. Mahmoud et al., (2004) investigate anaerobic sewage treatment in a one-stage UASB reactor and a combined UASB-Digester system. The UASB digester system represent an efficient technology for anaerobic (pre) treatment of 15ºC sewage at operating the digester at 35ºC, i.e. it provides average removal efficiencies for COD_t ,COD_{ss} , COD_{col} and COD_{dis} of 66%, 87%, 44% and 30%. Similarly Mahmoud (2008) studied the high strength sewage treatment one-stage UASB reactor in order to explain the influence of seasonal temperature fluctuations on the system performance over the first year of performance without inoculation and for HRT of 10h. In addition, a UASB-digester system was incorporated at 35ºC. As a result the performance was limited by the low temperature during winter time and the high strength and solids content. Khan et al., (2015) studied the long term performance of UASB reactor under controlled operation and maintenance at varying organic loads and investigate the sludge profile by validating the sludge blanket model. Study concluded that the performance of UASB reactor was shown to be very stable and robust, regardless of the temperature variation and the fluctuation in the influent characteristics. The removal of COD and BOD was not adversely affected during low temperature conditions since reactor was operated with well operation and maintenance. The effluent characteristics of pilot scale UASB reactor can be compared to that of properly operated and maintained full scale UASB reactors. The performance summary of pilot scale plants are presented in Table 2.1.

2.2 STUDIES ON FULL SCALE UASB REACTOR

Khan et al., (2011) investigate the aeration, hydraulic retention time (HRT), dissolved oxygen (DO), or oxygen transfer, on the quality of full scale UASB effluent. A 111 MLD sewage treatment plant in Ludhiana was monitored for 3 months. Treatment of effluent of pilot and full scale UASB operating at steady state was studied in an aeration-settling system. The fine pore submerged diffusers were used to aerate the effluent of UASB reactor under different operating conditions. Maximum removal efficiencies were achieved at 30 min HRT and dissolved oxygen (DO) of 5-6 mg/l. The performance of full scale surface aeration system was compared to the performance of pilot scale diffused aeration system. Pandey et al., (2014) reviewed the mechanisms of sludge reduction by UASB plants by using the cost analysis and environmental assessment and its practical approach to treat the water through UASB process. The study was conducted on 14 MLD UASB sewage treatment plant at Mirzapur. The Study concluded that UASB reactor was found to perform better and there is a considerable amount of biodegradable waste that is suitable for biogas production.

Another study was conducted by Walia et al., (2014) to check the performance of UASB based STPs in India. The UASB based STPs selected for study were 27 and 34 MLD at Noida, 56 and 70 MLD at Ghaziabad, and 38 MLD at Saharanpur. The installation of non-algal ponds reduced land requirement but remove only solids washed out of the UASB reactor. Khan et al., (2014) monitored the performance of 10 full scale UASB based STPs and investigate the performance of existing post treatment system. The summary of treatment of UASB reactors are presented in Table 2.2.

Table 2.1: Summary of studies on UASB pilot plant

T (˚C)	HRT (h)	Influent COD_t (mg/l)	Removal efficiency %	Biogass production (m^3/kg COD)	Methane production		SMA	Author
					Dissolved	Recovered		
8-40	8	461±393	65-85	0.18 – 0.22	N.A	N.A	40-60, 18-50 (ml CH_4/g VSS/d)	Khan et al., (2015)
35	10	1394	55	N.A	N.A	N.A	N.A	Mahmoud et al., (2008)
10		1137	32					
15	6	721	49	70 : 107	N.A	N.A	N.A	Mahmoud et al., (2004)
29	4	320	65	N.A	N.A	0.08-.2 (g CH_4/g VSS/d)	N.A	Kalogo et al., (2001)
25-13	4.7	15-595	69.4	N.A	0.16-0.27 Nlg^{-1} COD removed	<0.23 Nlg^{-1} COD removed	0.77 (Kg CH_4/kg VSS/d)	Uemura et al., (2000)
20	48		60-75	N.A	>60%	30-40%	N.A	Singh et al., (1998)

(N.A- Not Analysed)

Table 2.2: Summary of treatment of full scale UASB reactors

STP Location	Capacity (MLD)	Removal efficiency %			Post treatment	Removal efficiency %			Author
		BOD	COD	TSS		BOD	COD	TSS	
Ludhiana	111	66	58	43	Final Polishing Unit	67	80	52	Khan et al., 2011
Mirzapur	14	60	60	65	Polishing Pond	85	80	90	Pandey et al., 2014
Noida	27	53	41	59	Polishing Pond	27	46	27	Khan et al., 2014
Noida	34	79	51	54	Polishing Pond	10	41	34	
Saharanpur	38	60	55	60	Polishing Pond	43	45	57	
Agra	78	48	43	41	Polishing Pond	52	43	21	
Karnal	40	60	62	54	Polishing Pond	34	33	33	
Vadodra	43	62	75	70	Surface aeration + ASP	78	75	82	
Surat	100	47	42	40	Diffused aeration + ASP	86	81	65	
Balloke	152	59	55	49	Surface aeration + ASP	43	56	81	
Jamalpur	48	45	29	51	Surface aeration + ASP	54	86	71	

2.3 REVIEW ON UASBR TREATING DOMESTIC WASTEWATER

Khan et al., (2011) discussed the different systems for the treatment of UASB reactor effluent treating sewage. Additionally, a comparative review, an economic evaluation of some of the emerging options was conducted and based on the extensive review of different integrated combination, i.e. UASB-different aerobic systems, a treatment concept based on natural biological mineralization route recognized as an advanced technology to meet all practical aspects to make it a sustainable for environmental protection, resource preservation and recovering maximum resources. Based on the overall analysis of various post treatment systems, it was concluded that there is no ideal system applicable to all conditions. Each situation must be analysed individually, with the constant concern of incorporating the local specialties in the stage of investigation and decision. It can be said that the UASB system followed by an aerobic system can be the ideal concept for feasible and sustainable environment protection in a decentralized sanitation with resource recovery.

Dhote et al., (2012) review the use of wastewater treatment technologies to remove contaminants from wastewater such as halogenated hydrocarbon compounds, heavy metals, dyes, pesticides, and herbicides, which represent the main pollutants in wastewater and also review the various options that may be employed in the treatment, recovery and reuse of wastewater. Natural treatment technologies are considered viable because of their low capital costs, ease of maintenance, their potentially longer life-cycle and their ability to recover a variety of resources including: treated effluent for irrigation, organic humus for soil amendment and energy in the form of biogass.

Pandit et al., (2013) review the current research trends in wastewater and concluded that the efficient and proper wastewater purification processes thus can reduce the health related concerns associated with wastewater recycling. The performance efficiency of treatment plant depends on proper design and construction and also on good operation and maintenance. UASB applications for wastewater like sugar industry, distillery, dairy industry, slaughter house and high strength municipal wastewater was discussed by Powar et al., (2013) and removal efficiency of COD (Chemical Oxygen Demand) for various organic loading and HRT was studied.

Quaff et al., (2014) review the development of anaerobic process and technology for treatment of domestic wastewater. Treatment of sewage and applications of UASB reactor in India was studied by author.

2.4 OBJECTIVES

The objectives of the study are:

i. To assess the performance of full scale UASB reactor w.r.t COD removal for domestic wastewater under low temperature (psychrophilic) conditions.
ii. Biochemical Methane Potential (BMP) of domestic wastewater under low temperature conditions.
iii. Specific Methanogenic Activity (SMA) of sludge from full scale UASB reactor.

CHAPTER 3

MATERIALS AND METHODS

After fixing the objectives of the study, an experimental programme was conducted in two phases. Phase I consisted of full scale reactor studies while phase II consisted of batch studies.

3.1 FULL SCALE REACTOR STUDIES

3.1.1 Experimental Materials and Methods for reactor Studies

3.1.1.1 Full scale UASB reactor

An influent and effluent sample of UASB reactor of sewage treatment plant was collected to perform the experiments to calculate various parameters. The full scale UASB reactor is 32m long, 24m wide and 6m deep and is made up of RCC.

3.1.1.2 Analytical Procedures

Parameters considered for study of full scale UASB reactor experiment were analysed as per Standard Methods for the Examination of water and Wastewater (APHA, AWWA and WEF, 22nd Edition, 2012). The analytical techniques adopted are presented in Table 3.1. The frequency of analysis of various parameters in full scale reactor studies is given in Table 3.2. Grab sample was taken in each month for testing (Plate 3.1).

Plate 3.1: Grab sampling of domestic wastewater at Lalpani STP.

Table 3.1: Analytical Techniques adopted for the determination of Various Parameters

S.No.	Parameter	Method
1.	pH	Digital pH meter
2.	BOD	5 Day BOD Test
3.	COD	Open reflux
4.	Alkalinity	Titration method
5.	TS	Oven dry at 103-105ºC
6.	VS	Ignited at 550ºC

Table 3.2: Frequency of Analysis for Various Parameters

S.No.	Parameter	Frequency of analysis
1.	pH, Alkalinity	once a month
2.	BOD,COD	once a month
3.	TS,VS	once a month

3.1.1.2.1 pH Determination

pH of wastewater is determined by digital pH meter as shown in Plate 3.2.

Plate 3.2: Digital pH meter

3.1.1.2.2 Alkalinity Determination

Alkalinity of wastewater is defined as its acid-neutralizing capacity. Alkalinity parameter is used to control the various processes of wastewater. It is determined by Titration method. The set-up of titration method is shown in Plate 3.3.

Apparatus: Pipets, Flasks and Burets.

Reagents: Sodium sulphuric acid or hydrochloric (0.1N).

Indicators: Methyl Orange and Phenolphthalein (Plate 3.4).

Plate 3.3: Alkalinity test set-up

Plate 3.4: Indicators and Reagent

3.1.1.2.3 Biological Oxygen Demand Determination

Biological oxygen demand determines the relative oxygen requirements of wastewaters. The test measures the amount of oxygen consumed by microorganisms during a specific incubation period for biological degradation of organic matter. It is determined by 5-Day BOD test in which diluted wastewater is incubated at 20˚C temperature for 5 days. Initial DO and final DO is measured with DO probe. BOD_5 is determined by subtracting initial and final DO and multiply by dilution factor.

Apparatus: Incubation bottles (300mL) (Plate 3.5), Digital DO Meter (Plate 3.6), Incubator (Plate 3.7).

Plate 3.5: BOD Bottles

Plate 3.6: Digital DO meter

Plate 3.7: Incubator

3.1.1.2.4 Chemical Oxygen Demand Determination

The Chemical oxygen demand determines the amount of pollutant present in wastewater. It determines the amount of chemical oxygen consumed by microorganisms. COD is determined by Closed Reflux, Titrimetric Method.

Apparatus: Digestion vessels, Digester (Plate 3.8)

Reagents: Standard potassium dichromate solution (Plate 3.9), Sulphuric acid reagent, Standard Ferrous ammonium sulphate (FAS) (0.1N).

Indicator: Ferroin indicator solution (Plate 3.10).

Plate 3.8: Digester

Plate 3.9: Standard potassium dichromate

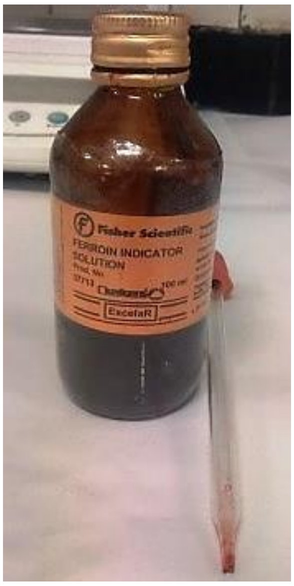

Plate 3.10: Ferroin Indicator

3.1.1.2.5 Solids Determination

Solids are present in two forms in wastewater, i.e., in suspended form and in dissolved form. Solids affect effluent quality in many ways. Total solids are the material residue left in the vessel after evaporation of the sample and drying in oven at 103˚C - 105˚C temperature. Total solids include suspended solids which are defined as the portion of total solids retained by filter paper and dissolved solids which pass through the filter paper.

1. Total Solids

Sample is evaporated and dried to constant weight in an oven at 103˚C - 105˚C temperature. The increase in weight of dish represents the total solids.

Apparatus: Oven (Plate 3.11), Evaporating dishes, Desiccator (Plate 3.12), Analytical balance (Plate 3.13) and Graduated cylinder.

Plate 3.11: Oven

Plate 3.12: Desiccator

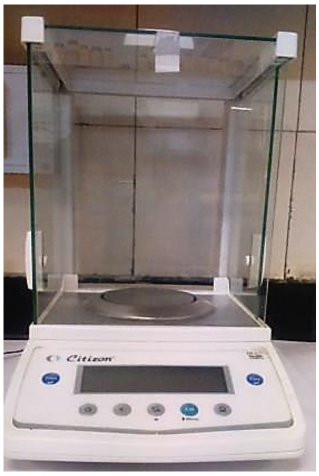

Plate 3.13: Analytical Balance

2. Dissolved Solids

The filtrate left during suspended solids filtration is evaporated in an oven to a constant weight. The increase in dish weight represents the dissolved solids.

3. **Volatile Solids**

The residue from section 1, 2 and 3 is ignited in Muffle furnace to constant weight at 550ºC. The weight loss on ignition represents the volatile solids and remaining solids are fixed solids. Apparatus: Oven, Muffle furnace (Plate 3.14), Crucibles (Plate 3.15), Desiccator, Analytical balance and Graduated cylinder.

Plate 3.14: Muffle furnace

Plate 3.15: Crucible

The organic solid concentration in reactor is obtained from the weight loss during the ignition of total solids. The amount of volatile solids in wastewater indicates the presence of amount of organic matter.

3.2 BATCH STUDIES

Batch studies were conducted at different temperatures in serum bottles of 125 mL capacity with rubber caps and aluminium seals according to the procedure given by Owen et al., (1979).

Following batch studies were conducted:

1. Biochemical methane potential (BMP) of domestic wastewater under low temperature conditions.
2. Specific methanogenic activity (SMA) of sludge for full scale UASB reactor at 20ºC.

3.2.1 Experimental Materials and Techniques in Batch Studies

3.2.1.1 Inocula

For BMP of wastewater UASB sludge was used as Inocula. Sludge was taken from an anaerobic UASB reactor of sewage treatment plant (19.35 MLD capacity plant treating sewage) at Lalpani, Shimla, Himachal Pradesh, India. Characteristics of the sludge are presented in Table 3.3.

Table 3.3: Characteristics of sludge used as Inocula in batch studies

Parameters	**UASB sludge**
TSS, g/L	50.4
VSS, g/L	32.7
% VSS in TSS	64.8
Colour of sludge	Black

3.2.1.2 Wastewater

UASB influent (from Lalpani STP) was used in batch studies to access its biodegradability.

3.2.1.3 Preparation of Bottles

Serum bottles were thoroughly washed in tap water. The bottles were dried and placed in an incubator. Total liquid volume (containing sample and Inocula) of 100 mL was used in order to maintain appropriate liquid-to-void ratio for precision and accuracy of results. A serum cap was placed after the bottle was filled to the appropriate volume while simultaneously removing the oxygen from the bottle by flushing nitrogen gas in it. The stoppers were fitted with an aluminium crimp seal.

3.2.1.4 Gas Measurements

Glass syringe was used to measure gas volume present in headspace. The plunger was lubricated with water and serum bottles were shaken properly before gas volume measurements were made. The gas volume measurement from serum bottles using glass syringe is shown in Figure 3.1.

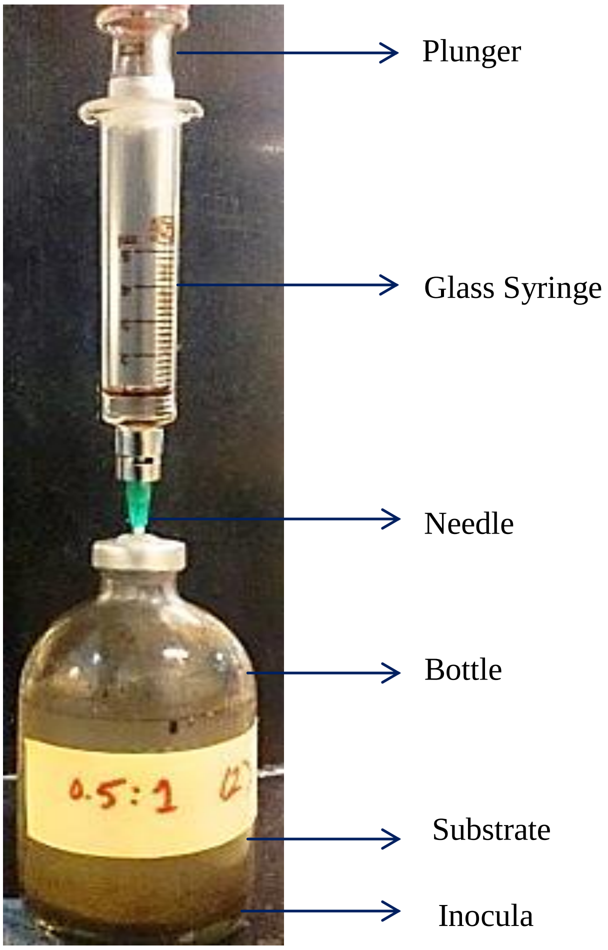

Figure 3.1: Gas volume measurement from serum bottles using glass syringe

3.2.2 Biochemical Methane Potential

Biochemical methane potential (BMP) is one of the most common parameters for assessing the biodegradability of waste. Each serum bottle was filled with a feed containing wastewater and sludge in varying amounts. The various F/M ratios included 4, 1.3, 0.35 and 0.1. Set of BMP bottles is shown in Plate 3.15.

3.2.3 Specific Methanogenic Activity

Specific Methanogenic Activity (SMA) was conducted in order to assess the potential of the anaerobic granular sludge from full scale UASB reactor treating domestic wastewater. Each

SMA study consisted of 15 bottles with experiments performed in duplicate. The bottles were charged with different substrate to a sludge ratio of 2, 1 and 0.5. Sodium acetate is used as substrate. Set of SMA bottles is shown in Plate 3.16.

Plate 3.15: BMP bottles

Plate 3.16: SMA bottles

CHAPTER 4

RESULTS AND DISCUSSION

4.1 CONTINOUS REACTOR

Grab samples were analysed from a full scale UASB based STP for Temperature, pH, Alkalinity, Dissolved Solids (DS), Suspended Solids (SS), Volatile Solids (VS), BOD and COD. Samples were collected once in a month during winter i.e. November – March. The results of the above parameters are tabulated in APPENDIX I and discussed briefly below.

4.1.1 Temperature

The temperature of municipal wastewater in UASB inlet and outlet, varied between 10ºC and 18ºC. Temperature less than 20ºC is considered as psychrophilic temperature, which slows down the rate of anaerobic digestion of wastewater (Bogte et al., 1993; Van Haandel and Lettinga, 1994). However, such extreme temperature usually last only for the winter months, i.e., January to March. The temperature during winter months in inlet and outlet of UASB reactor is shown in Table 4.1.

Table 4.1: Temperature in inlet and outlet of UASB reactor

Month	Inlet (ºC)	Outlet (ºC)
November	18	17
December	10	11
January	15	15
February	16	16
March	14	14

4.1.2 pH

pH is hydrogen ion concentration and is an important parameter in treatment of biological units. The pH-value is also important because at high pH, ammonia (NH_4) dissociates to NH_3 which inhibits the growth of the methane producing bacteria (Tilley et al., 2014). The pH value fluctuations in the influent and effluent of the UASB reactor were monitored over time and the average pH values (during the study period) in the inlet and outlet were 7.3 and 7.2, respectively (Table 4.2). The drop in pH value in effluent is due to the decomposition of organic matter. Monitoring of pH value in the anaerobic reactor is decisive and can be helpful in determining abnormalities of a system. In present study, the pH value of the treated domestic wastewater (effluent) was in the range of 7.1-7.25, maintaining nearly the optimum level for the methanogenic activity. Methane producing bacteria work better in a pH range of 6.7-7.4.

Table 4.2: Influent and effluent pH values

Month	Temperature °C	Influent pH value	Effluent pH value
November	18	7.2	7.1
December	10	7.28	7.24
January	15	7.3	7.25
February	16	7.31	7.2
March	14	7.5	7.2

4.1.3 Colour and odour

Fresh domestic sewage is greyish in appearance, but as time passes it becomes dark in colour with a pronounced smell due to microbial activity. By physical observations, the colour of the influent and effluent appeared grey.

4.1.4 Alkalinity

Alkalinity of wastewater determines the acid neutralizing capacity of wastewater. More alkalinity was observed in outlet than inlet of UASBR (Table 4.3). The average alkalinity of influent and effluent were (391 ± 16) mg/L and (453 ± 32) mg/L. From these results it can be concluded that sufficient amount of alkalinity is present in a reactor to resist major change in pH.

Table 4.3: Influent and effluent Alkalinity of reactor

Month	Temperature	Alkalinity_{in}	Alkalinity_{out}
November	18	382	400
December	10	370	462
January	15	410	486
February	16	389	460
March	14	405	456

4.1.5 Solids

The solids are classified into suspended solids, dissolved solids and volatile solids. Determination of volatile or organic fraction of solids is necessary as this constitutes the load on biological treatment units when sewage is disposed-off. Dissolved inorganic fraction determines the applicability of sewage for land irrigation or any other reuse (Choksi et al., 2015). The average concentrations of suspended solids in the influent and effluent of the UASB reactor were 1160mg/l ± 410 and 490mg/l ± 434 (Figure 4.1). The solids removal efficiency throughout the treatment process in different months of the winter season was 9-82%. The results provided in Table 4.4 shows different removal efficiencies in the inlet and outlet of the reactor. The maximum removal efficiency was 82%, which corresponds to an influent SS of 1126mg/L and the minimum SS removal efficiency was 9%, which corresponds to an influent SS of 796 mg/L. Performance of reactor was not good in February because UASB reactor was not running at that day.

Table 4.4: Influent and effluent suspended solids concentrations and removal efficiencies

Month	Temperature °C	SS_{inf} (mg/L)	SS_{eff} (mg/L)	Removal efficiency %
November	18	1863	1150	38
December	10	1126	205	82
January	15	1009	186	81
February	16	796	722	9.0
March	14	1009	186	81

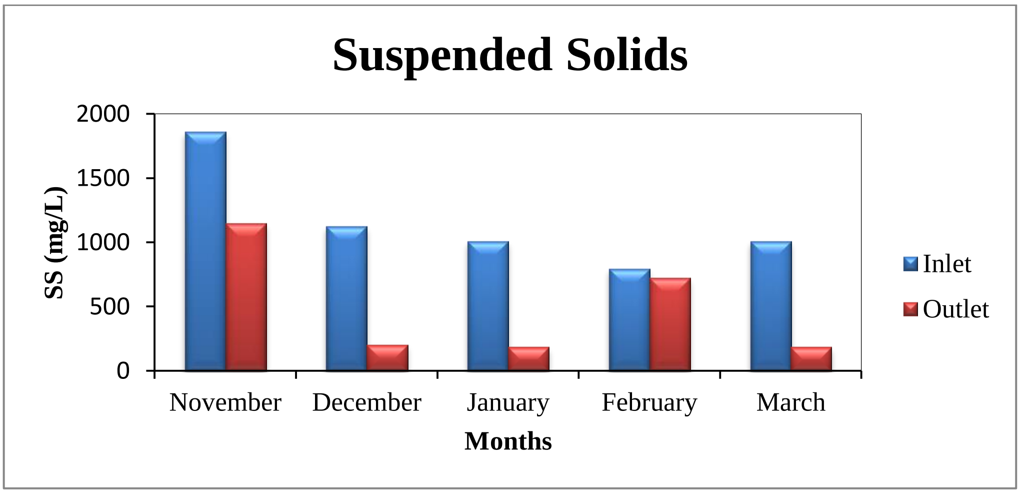

Figure 4.1: Suspended solids variations at inlet and outlet of UASB reactor

The inconsistency in performance of UASB was due to fluctuations in influent characteristics, improper desludging and poor operation and maintenance (Khan et al., 2014). Excess sludge accumulation and increase of upflow velocity more than 0.35m/h are the major factors responsible for increase and decrease in suspended solids concentrations (Lew et al., 2003; Walia et al., 2014). The influent and effluent mean concentrations of dissolved solids were 588 mg/L ± 27 and 618 mg/L ± 28. The results provided in Table 4.5 shows the different values of dissolved solids in inlet and outlet. The experimental results show that the concentration of DS in outlet is more than in inlet except in month of December. Figure 4.2 shows the variation in DS concentrations during different months.

Table 4.5: Influent and effluent dissolved solids concentrations

Month	**Temperature °C**	**DS_{inf} (mg/L)**	**DS_{eff} (mg/L)**
November	18	611	668
December	10	617	608
January	15	570	605
February	16	586	607
March	14	554	603

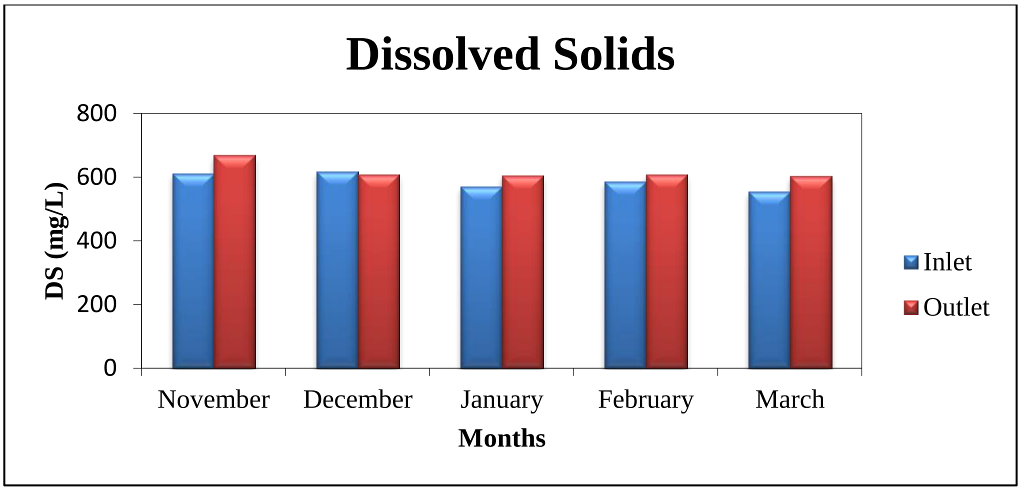

Figure 4.2: Dissolved solids variations at inlet and outlet of UASB reactor

Increase in dissolved solids concentration (Figure 4.2) is due to poor UASB performance, granulation failure and incomplete consumption of dissolved organic matter by microorganisms. It may be due to high hydraulic loading rate or increase in sludge level. The organic solid concentration in reactor is obtained from the weight loss during the ignition of total solids. The amount of volatile solids in wastewater indicates the presence of amount of organic matter. The average concentrations of volatile solids in influent and effluent of reactor were 823 mg/L ± 165 and 418 mg/L ± 211 during the winter season. The results provided in Table 4.6 shows the variation of VS in inlet and outlet. The removal efficiency varied between 8 to 67%. Figure 4.3 shows the variation of VS in the inlet and outlet of the reactor. Reduction in volatile solids was clearly an indication of active biomass growth in reactor. Similar observations have been reported elsewhere in the literature (Rizvi et al., 2015). It also signifies the production of biogas.

Table 4.6: Influent and effluent volatile solids concentrations and removal efficiencies

Month	Temperature °C	VS_{inf} (mg/L)	VS_{eff} (mg/L)	Removal efficiency %
November	18	584	195	67
December	10	1046	360	65
January	15	829	511	38
February	16	798	734	8.0
March	14	860	288	66

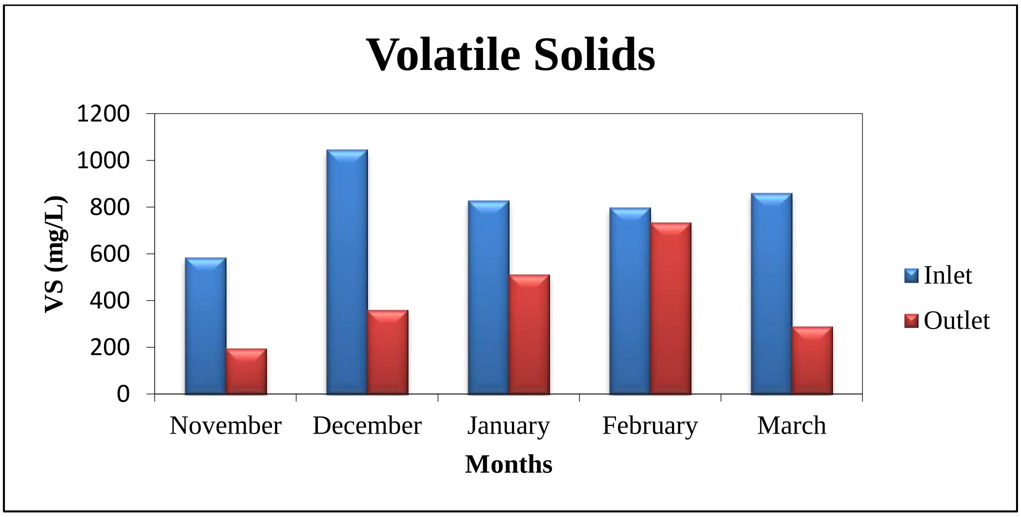

Figure 4.3: Volatile solids variations at inlet and outlet of UASB reactor

4.1.6 BOD and COD removal

BOD and COD are two parameters used to access the strength of organic matter present wastewater. The average concentration of COD in the influent and effluent of the UASB reactor were found to be 633 mg/L ± 110 and 489 mg/L ± 137 respectively. 13 to 42% of COD reduction was observed. The results provided in Table 4.7 shows the COD removal at different temperature.

Table 4.7: Influent and effluent COD concentrations and removal efficiencies

Month	Temperature °C	C_{inf} (mg/L)	C_{eff} (mg/L)	Removal efficiency %
November	18	550	320	42
December	10	736	576	22
January	15	536	428	20
February	16	768	672	13
March	14	576	448	22

The experimental results show that low temperature has significant influence on COD removal. COD removal rates were found to be independent of the influent concentrations. The maximum COD removal was 42%, which corresponds to an influent COD of 550 mg/L at 18ºC temperature and the minimum COD removal rate was 13% corresponding to an influent COD of 768 mg/L at 16ºC. The variations of COD during winter months are shown in Figure 4.4. A drop of 8ºC resulted in around 50% reduction in COD during November – December.

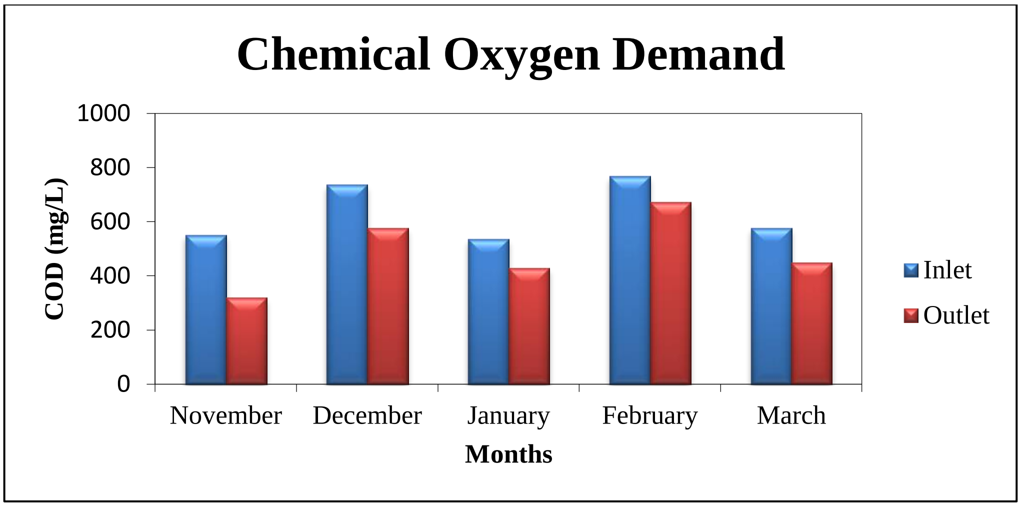

Figure 4.4: COD variations at inlet and outlet of UASB reactor

The average BOD concentrations in the influent and effluent of the UASB reactor were 365 mg/L ± 76 and 275 mg/L ± 96 respectively. The results provided in the Table 4.8 shows the effect of low temperature on removal of BOD.

Table 4.8: Influent and effluent BOD concentrations and removal efficiencies

Month	Temperature ºC	C_{inf} (mg/L)	C_{eff} (mg/L)	Removal efficiency %
November	18	286	184	36
December	10	422	318	25
January	15	280	230	18
February	16	437	424	3
March	14	401	221	22

The BOD removal efficiency was 3 to 36% under psychrophilic conditions. The maximum BOD removal efficiency was 36% which corresponds to an influent BOD of 286 mg/L at 18ºC. The variations of BOD are shown in Figure 4.5. Low removal efficiency under psychrophilic conditions may be due to incomplete sludge granulation and inadequate volume of settled solids and biomass; therefore, slows down the hydrolysis and reduce the methanogenic activity of sludge microorganisms (Hulshoff Pol, 1989; Lehtomaki et al., 2008; Lettinga et al., 2008; Van der Last and Lettinga, 1992). The BOD removal efficiency and the quality of the effluent depend on the retention time and temperature. The reduction of BOD simultaneously decreases the coliforms (Der Steen et al., 2000).

Low removal efficiency of BOD and COD was due to improper operation and maintenance (O&M) of the UASB reactor. UASB reactor operation requires various activities which are necessary for better performance of a reactor. Activities like process performance monitoring, collection of data, collection of influent and effluent samples for testing and collection of sludge samples etc. Monitoring of sludge and its profile inside the reactor is one of the major operational activities related to UASB plants (Khalil et al., 2008). Maintenance of UASB plant includes removal of floating layers inside the gas dome at least once in six months, cleaning and repairing of gutters, V- notch weir plates, baffles and feeding boxes, checking of the level of the overflow weirs yearly, removal of floating scum material at the top of the water surface of the reactor once a day and cleaning of chocked feeding pipes (Lalpani STP O/M Manual).

BOD/COD ratio is the most important parameter that identify whether the sewage is biodegradable or non-biodegradable and to what extent (Pandit et al., 2013). The BOD/COD ratio of Lalpani sewage was around 0.55 which means that it is a biodegradable waste. The

BOD/COD ratio of influent and effluent of UASB reactor for each experiment is provided in Table 4.9.

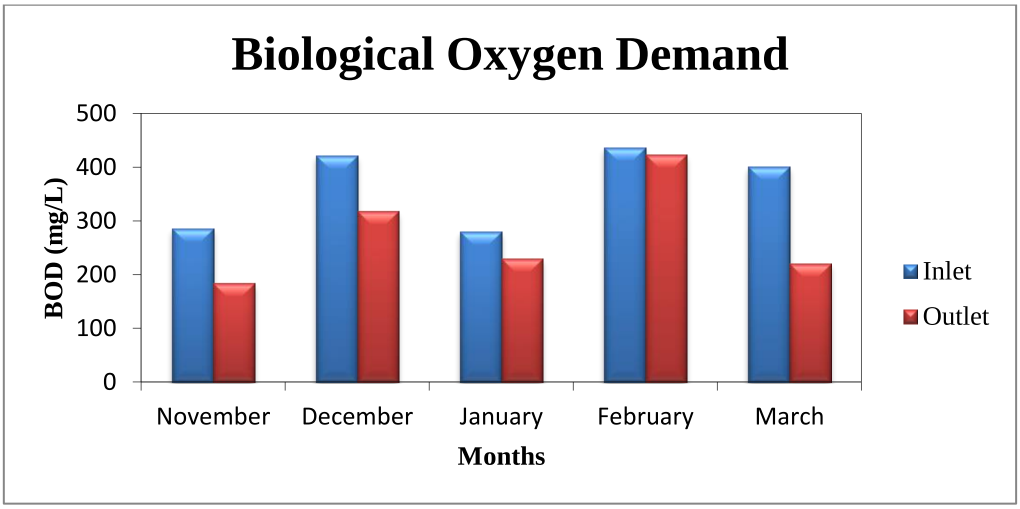

Figure 4.5: BOD variations at inlet and outlet of UASB reactor

Table 4.9: BOD/COD ratio for Influent and effluent of UASB reactor

Month	Temperature °C	BOD/COD Inf.	BOD/COD Eff.
November	18	0.52	0.57
December	10	0.57	0.55
January	15	0.52	0.53
February	16	0.56	0.63
March	14	0.60	0.49

4.2 BATCH STUDIES

Results of batch biodegradability studies are influenced by the methodology adopted, source of inoculum, storage conditions, adaptability and activity of sludge. The main factor which influences the degradability rate is the ratio of substrate to sludge biomass i.e. F/M ratios (0.1, 0.35, 1.3 and 4). Two sets of experiments were conducted:

Set I: Temperature controlled condition (19°C)

F/M ratio 0.1, 0.35, 1.3 and 4

Set II: Temperature uncontrolled condition (0-26°C)

F/M ratio 0.1, 0.35, 1.3 and 4

4.2.1 Biochemical Methane Potential of Sewage Wastewater

Anaerobic biodegradability potential of a substrate is expressed in terms of net cumulative CH_4 production as a percentage of the theoretical CH_4 production calculated from the stoichiometry of the substrate. In mixed substrate, the expected CH_4 production is calculated based on the following reaction:

$$CH_4 + O_2 \longrightarrow CO_2 + 2H_2O$$

$$16 \quad\quad 64$$

One mole of CH_4 at standard temperature and pressure (STP: 0ºC and 1atm) occupies a volume of 22.4L. Thus

$$64 \text{ g COD stabilized} = 22.4 \text{ L of } CH_4$$

i.e., $1 \text{ g COD stabilized} \sim 0.35 \text{ L } CH_4 \sim 1 \text{ g } CH_4\text{-COD}$

4.2.1.1 Cumulative Biogas and Methane Production

For each F/M ratio (0.1, 0.35, 1.3 and 4) of BMP determination, cumulative CH_4 production was monitored. Domestic wastewater from the inlet of UASB was used as substrate. However, no macro or micro nutrients were added. BMP determinations were made under different temperature conditions. With UASB sludge as inoculum, biogass was observed for 28 days at 19ºC in controlled conditions for first round of experiment. The cumulative biogas and methane production for second round of experiment was performed for 31 days at ambient temperature (0-26ºC). Net cumulative biogas production was calculated after deducting background gas production due to biomass (blank samples without substrate). The numerical values of biogas and methane are incorporated in APPENDIX II.

For first set of experiment the biogas production was very low during the initial period. But with time a rise was observed in biogas production. Figure 4.6 illustrate the daily cumulative biogas production for first round of experiment. The reason for low biogas production can be due to psychrophilic conditions (<20ºC) compared to mesophilic conditions (>35º). Net cumulative biogas production was calculated after deducting background gas production and the net cumulative biogas is shown in Figure 4.7. It was noticed from Figure 4.6 that the total gas production for F/M ratio 0.1 was more than other F/M ratios. But net gas production for F/M ratio 4 was more than 0.1. A negative gas production was observed during experiment which indicates that the gas production was more from sludge as compared to substrate. It was assumed that 70% of methane is present in the total biogas. Cumulative methane production profiles from degradation of substrate at varying F/M ratios are shown in Figure 4.8 (APPENDIX II).

Stoichiometrically, the CH_4-COD for F/M ratio 4, 1.3, and 0.35 were expected to 217 mg (76 mL CH_4), 223 mg (78 mL CH_4) and 200 mg (70 mL CH_4) respectively. However, the observed values are 93.5 mg (32.7 mL CH_4), 81.3 mg (28.4 mL CH_4) and 33 mg (11.5 mL CH_4) respectively. The reasons for the observation recorded could be the gas leakage during measurement of gas and entrapment of the gas by the granules. It can also be conclude that some portion of dissolved methane has escaped from the effluent. Under low F/M ratio i.e. 0.1, the biogas contributed by the biomass was significant. That means, increase in sludge concentration in reactor will decrease the efficiency of reactor.

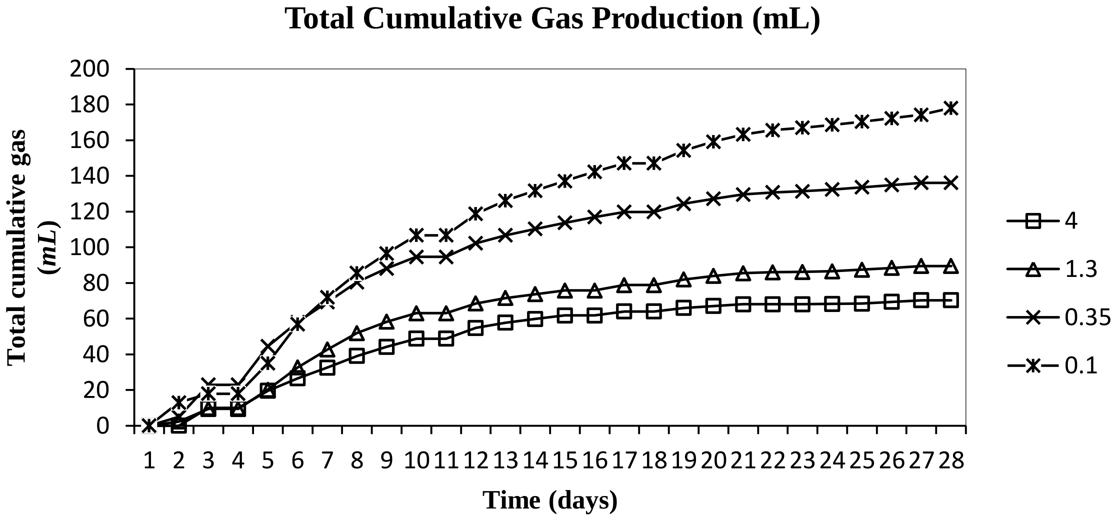

Figure 4.6: Total cumulative biogas production from different F/M ratios (0.1- 4) at 19°C

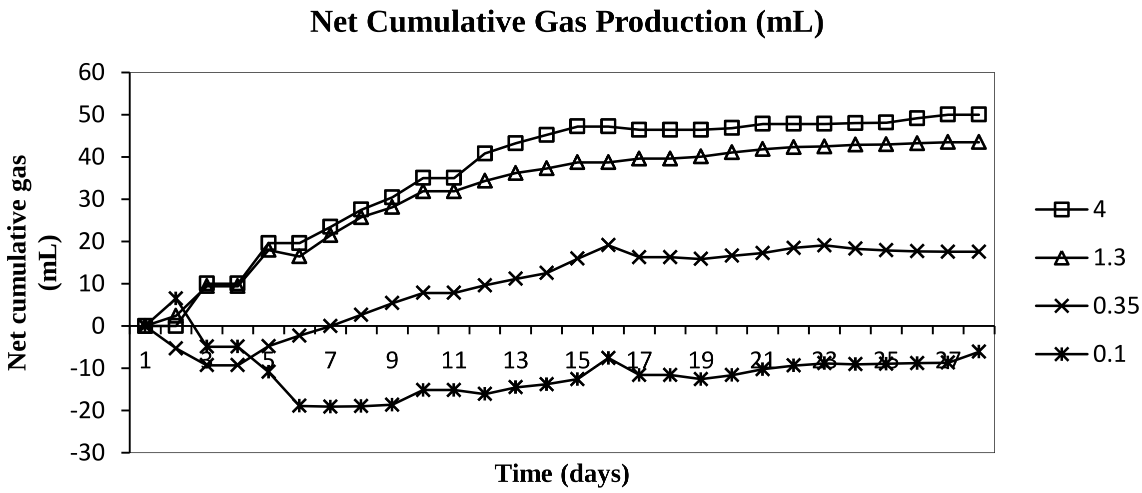

Figure 4.7: Net cumulative gas production from different F/M ratios (0.1- 4) at 19°C

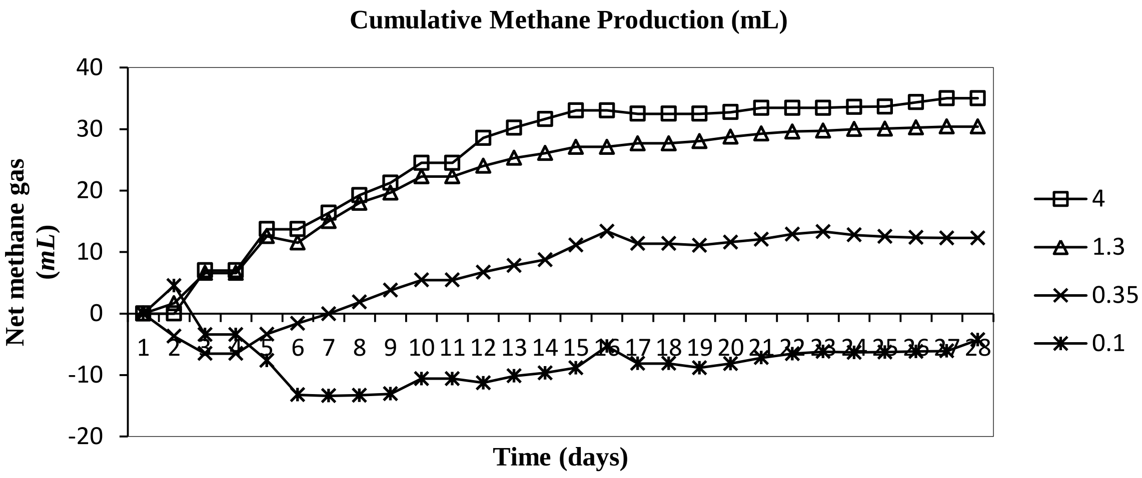

Figure 4.8: Cumulative methane production from different F/M ratios (0.1- 4) at 19°C

For second set of experiment, which was conducted at ambient temperature (0-26°C) (Figure 4.9), the biogas production was low during the initial period of 7 days. However, biogas production increased during the remaining period of experimental study. Figure 4.10 illustrate the daily cumulative biogas production for second set of experiment and Figure 4.11 and Figure 4.12 represent the net cumulative biogas production and methane production. The data of biogas and methane production are incorporated in APPENDIX II.

Stoichiometrically, the expected CH_4-COD for F/M ratios 4, 1.3, 0.35 was estimated to be 154 mg (54 mL CH_4), 174 mg (61 mL CH_4), and 885 mg (310 mL CH_4) respectively. However, the observed values are 59 mg (20.61 mL CH_4), 51 mg (17.89 mL CH_4), 26 mg (9.2 mL CH_4) respectively.

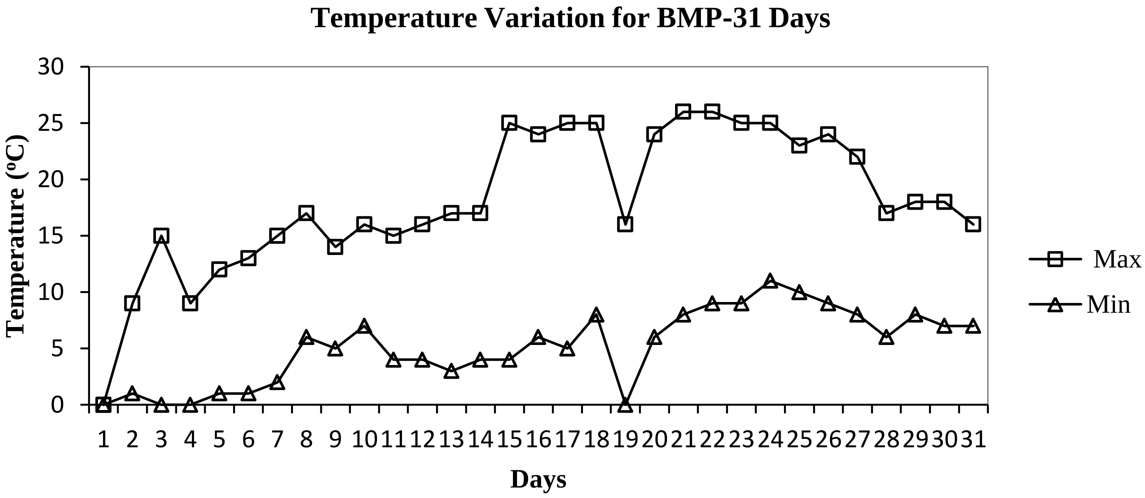

Figure 4.9: Temperature variation for BMP - 31 day

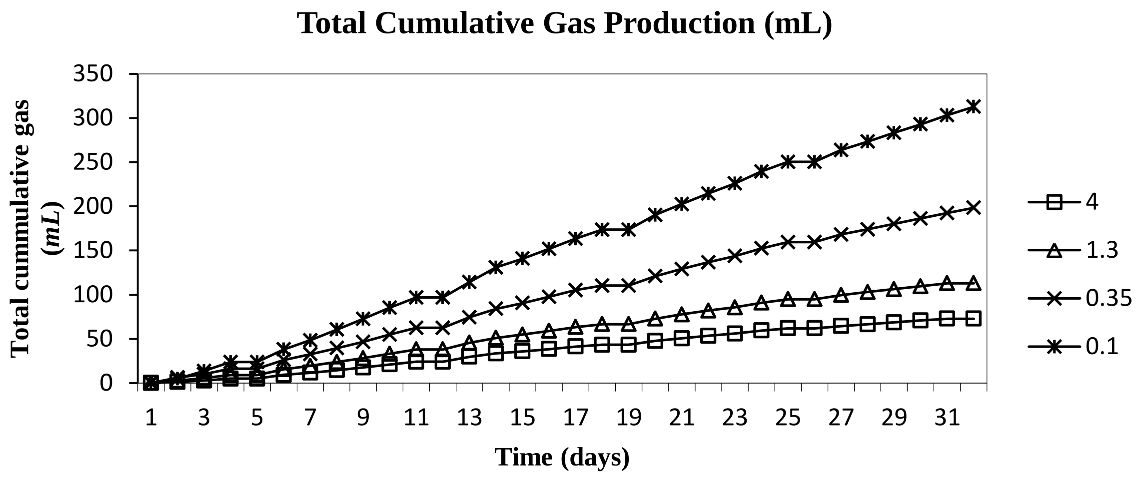

Figure 4.10: Total cumulative biogas production from different F/M ratios (0.1- 4) at ambient temperature

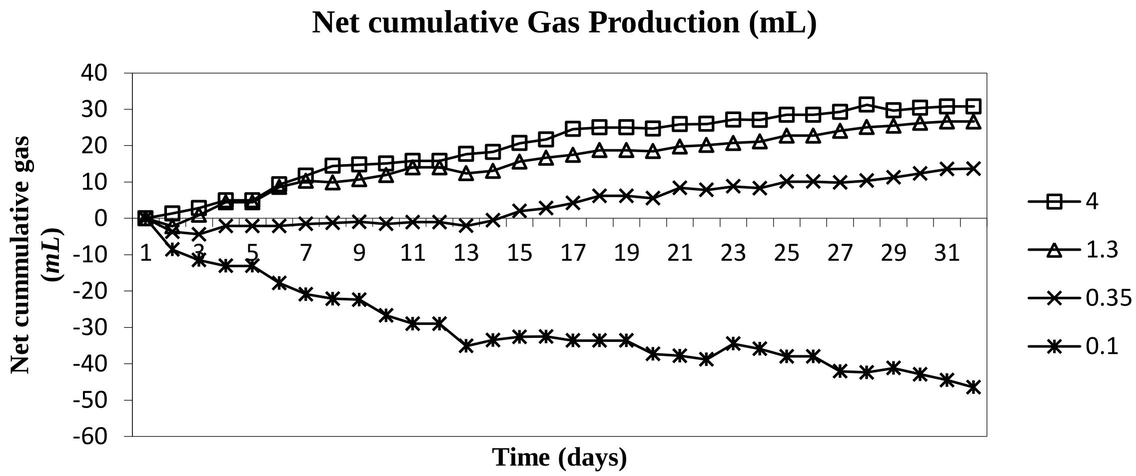

Figure 4.11: Net cumulative gas production from different F/M ratios (0.1- 4) at ambient temperature

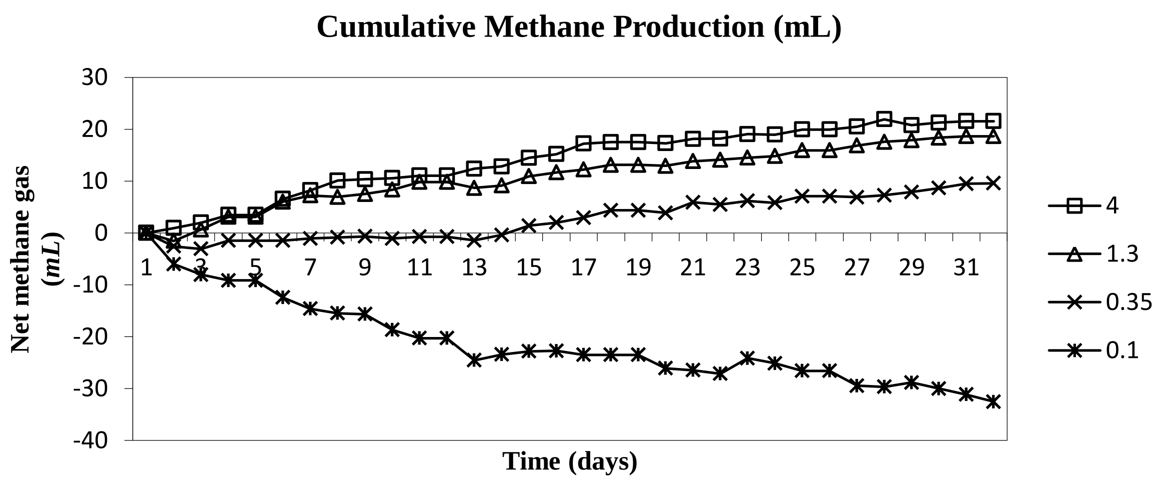

Figure 4.12: Cumulative methane production from different F/M ratios (0.1- 4) at ambient temperature

It was observed that total gas production was more during ambient conditions than controlled conditions, but cumulative methane production was found to be more in controlled conditions. This shows that temperature plays an important role in gas production. Frequent fluctuations in temperature affect the methanogenic phase by affecting the methane forming bacteria. Thus, the operating temperature of reactor is required to maintain as constant.

4.2.1.2 Influence of F/M Ratio on Anaerobic Degradation

The effect of F/M ratio on biogas production was studied for first and second set of experiment. Cumulative methane production at F/M ranging from 0.1 to 4 was monitored for controlled and uncontrolled conditions. The experimental results illustrated in Table 4.10 and 4.11 shows that with increase in F/M ratio the production of methane also increases. Negative gas production was also observed which may be because of more background gas production. This negative gas production may cause poor removal of substrate which can affect the efficiency of reactor.

Table 4.10: Cumulative gas production for first set of experiment

F/M ratio	Temperature °C	Total biogas production (mL)	Net biogas production (mL)	Methane production (mL)	Methane production at STP (mL)
4	19	70.30	50.00	35.00	32.74
1.3	19	89.46	43.46	30.40	28.45
0.35	19	136.15	17.55	12.30	32.85
0.1	19	178.00	-6.10	-4.30	-3.99

4.2.1.3 BMP

BMP is estimated by calculating methane production in g CH_4-COD and divided by g COD fed. BMP for different F/M ratios for first and second set of experiment are given in Table 4.12 and 4.13. BMP indicate degradability and methane production per unit of COD and it can be used as index of the anaerobic biodegradation potential for maximum quantity of methane produced per gram VSS added (Hussain et al., 2015). From Table 4.12 and 4.13, it was observed that the BMP value is more for F/M ratio 4. F/M ratio 4 has more gas production while other ratios have poor gas production. With decrease in F/M ratio the BMP value also decreases.

Table 4.11: Cumulative gas production for second set of experiment

F/M ratio	Temperature ºC	Total biogas production (mL)	Net biogas production (mL)	Methane production (mL)	Methane production at STP (mL)
4	0-26°C	72.80	30.77	21.5	20.61
1.3	0-26°C	112.90	26.63	18.64	17.89
0.35	0-26°C	198.35	13.65	9.55	9.20
0.1	0-26°C	312.60	-46.5	-32.5	-30.76

Table 4.12: BMP of different F/M ratios for first set of experiment

F/M ratio	Temperature ºC	Methane (g CH_4 COD) at STP	BMP (g CH_4 COD/COD_{fed})	BMP (mL CH_4/ g VSS)
4	19	0.093	0.28	137
1.3	19	0.081	0.21	80
0.35	19	0.033	0.04	20

Table 4.13: BMP of different F/M ratios for second set of experiment

F/M ratio	Temperature ºC	Methane (g CH_4 COD) at STP	BMP (g CH_4 COD/COD_{fed})	BMP (mL CH_4/ g VSS)
4	0-26°C	0.059	0.087	67
1.3	0-26°C	0.051	0.067	36
0.35	0-26°C	0.026	0.020	7

For better digestibility and methane production, the F/M ratio should be maintained properly so there must be enough food for microorganisms.

4.2.1.4 COD Mass Balance

The COD mass balance has been done by considering the following equation.

$$COD_{initial} = COD_{final} + VSS\ COD + CH_4\text{-}COD.$$

For first set of experiment, F/M ratio 4, the COD removal was 65% while 35% remained in final COD. Out of 65% COD removal, 28% of COD was converted to CH_4-COD and remaining 37 % COD has been equated to biomass COD. Similarly for F/M ratio 1.3 and 0.35 %COD removal is shown in Table 4.14.

Table 4.14: COD conversion for different F/M ratios for first set of experiment

F/M ratio	%COD Removal	% residual COD	% COD converted to CH_4-COD	% COD converted to Biomass COD
4	65	35	28	37
1.3	58	42	21	37
0.35	26	74	4	22

For second set of experiment, F/M ratio 4, the COD removal was 23% while 77% remained in final COD. Out of 23% COD removal, 9% of COD was converted to CH_4-COD and remaining 14 % COD has been equated to biomass COD. Similarly for F/M ratio 1.3 and 0.35 %COD removal is shown in Table 4.15.

Table 4.15: COD conversion for different F/M ratios for second set of experiment

F/M ratio	%COD Removal	% residual COD	% COD converted to CH_4-COD	% COD converted to Biomass COD
4	23	77	9	14
1.3	23	77	16	7
0.35	76	23	2	75

4.2.2 Specific Methanogenic Activity of Sludge

SMA determines the capability of sludge to produce methane by using a specific substrate. The SMA of sludge was determined at controlled temperature condition (20ºC) in a temperature controlled incubator. CH_4 generation was recorded through serum bottle technique to assess the degradability of substrate. Two sets of experiments were conducted:

Set I: SMA for 15 days at 20ºC

Set II: SMA for 33 days at 20ºC

For the first set of experiment, the daily cumulative gas production for each F/M ratio is shown in Figure 4.13 and net cumulative gas and cumulative methane production are shown in Figure 4.14 and Figure 4.15 (APPENDIX II). It was noticed from Figure 4.13 that the total gas production for F/M ratio 0.5 was more than ratios 1 and 2. But net gas production for F/M ratios 1 and 2 were more than 0.5. This was because of more background gas production.

Similarly, for second set of experiment, the daily cumulative gas production for each F/M ratio is shown in Figure 4.16 and net cumulative gas and cumulative methane production are shown in Figure 4.17 and Figure 4.18 (APPENDIX II). It was noticed from Figure 4.16 that the total gas production for F/M ratio 0.5 was more than ratios 1 and 2. But net gas production for F/M ratios 1 and 2 were more than 0.5. Figure 4.18 illustrate that methane production is more for F/M ratio 2 as compared to other ratios. This concludes that F/M ratio 2 is better than other two ratios for methane production.

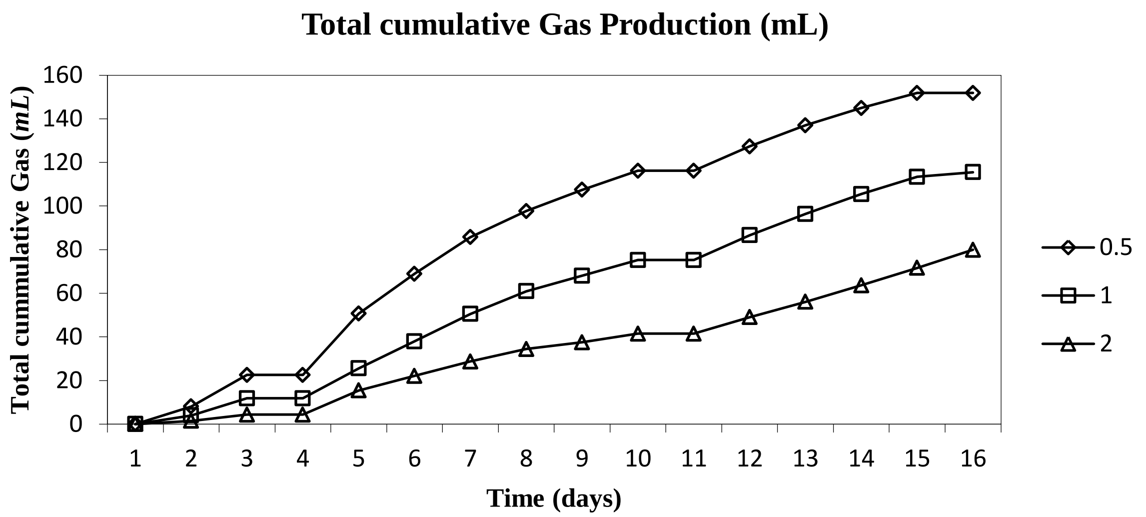

Figure 4.13: Total cumulative gas production from different F/M ratios (0.5-2) for first set of experiment

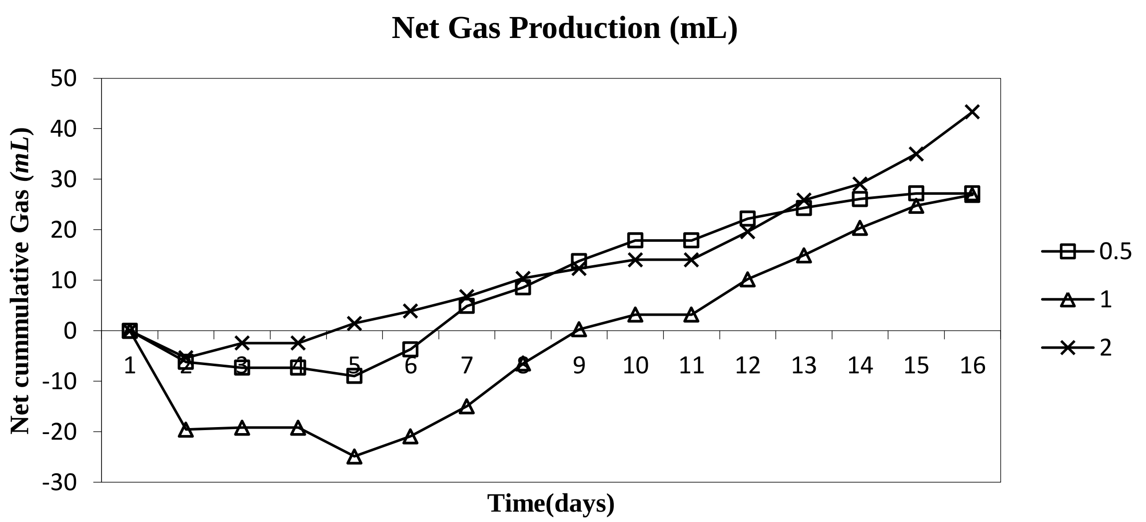

Figure 4.14: Net cumulative gas production from different F/M ratios (0.5-2) for first set of experiment

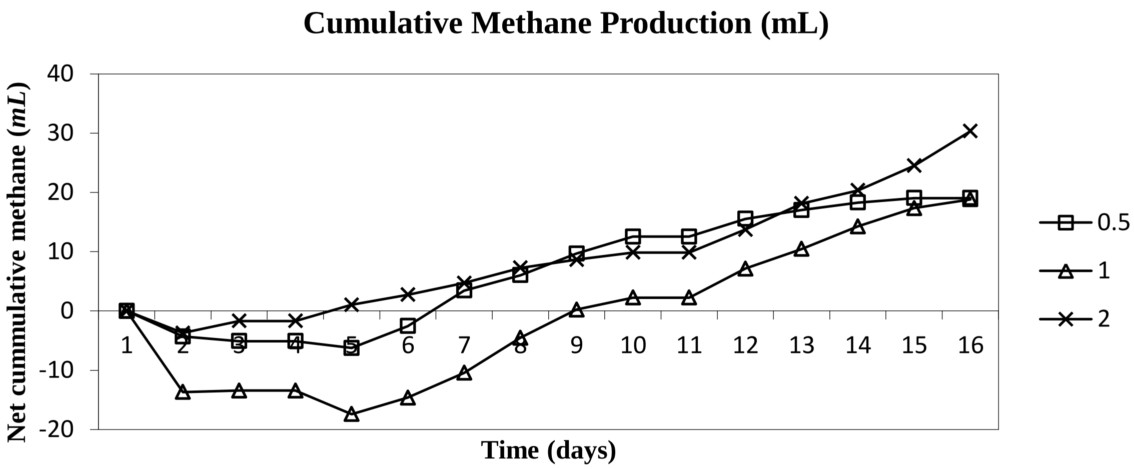

Figure 4.15: Cumulative methane production from different F/M ratios (0.5-2) for first set of experiment

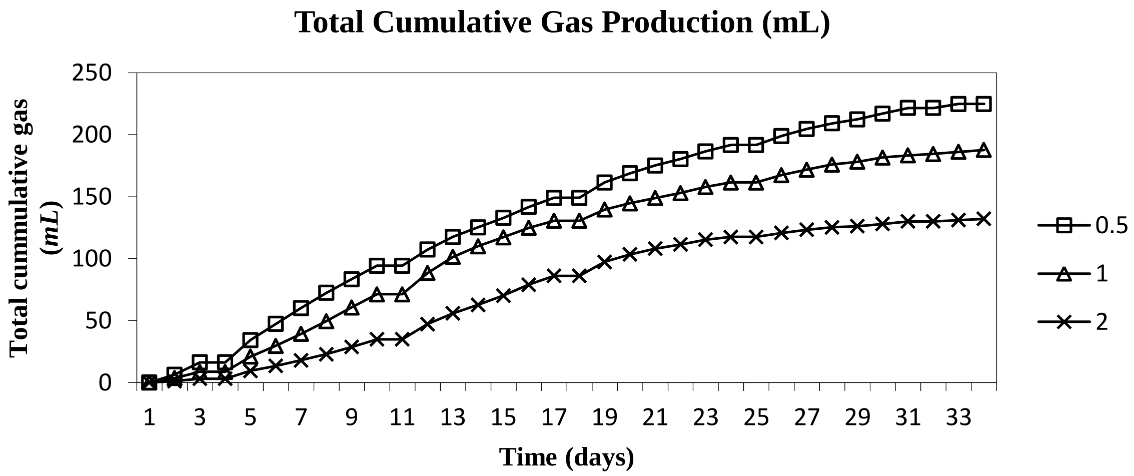

Figure 4.16: Total cumulative gas production from different F/M ratios (0.5-2) for second set of experiment

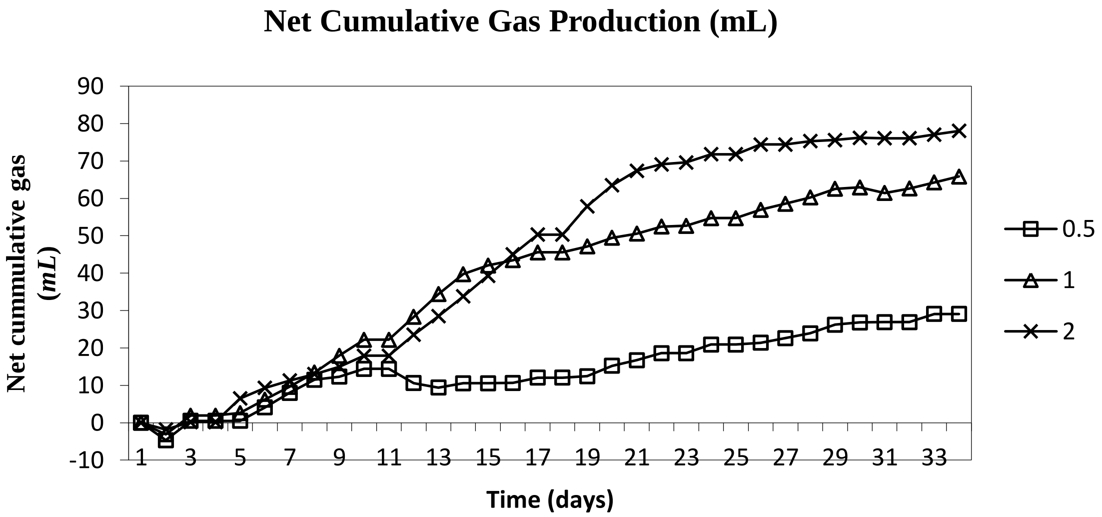

Figure 4.17: Net cumulative gas production from different F/M ratios (0.5-2) for second set of experiment

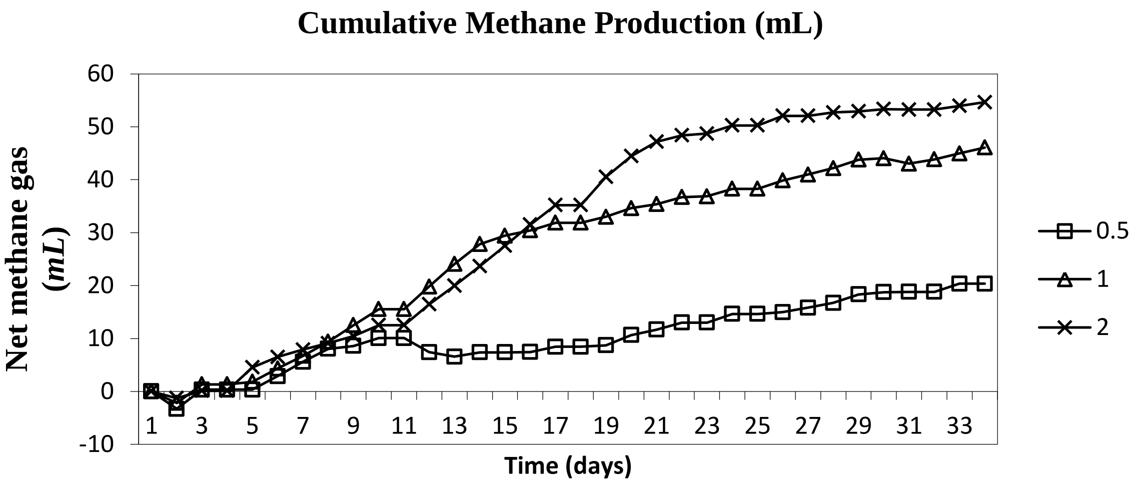

Figure 4.18: Cumulative Methane production from different F/M ratios (0.5-2) for second set of experiment

4.2.2.1 Influence of F/M Ratio on Anaerobic Degradation

The effect of F/M ratio on biogas production was studied for SMA test. The results of SMA experiments clearly indicate that F/M ratio has a great influence on biogas production. The experimental results showed in Table 4.16 and 4.17 shows that with increase in F/M ratio the production of biogas decreases but net biogas production increases. Negative gas production was also observed which may be because of more background gas production. This negative gas production may cause poor removal of substrate.

Table 4.16: Cumulative gas production for different F/M ratios (0.5-2) for first set of experiment

F/M ratio	Temperature ºC	Total biogas production (mL)	Net biogas production (mL)	Methane production (mL)	Methane at STP (mL)
2	20	80	43	30	28.27
1	20	115	26	18	17.55
0.5	20	152	27	19	17.70

Table 4.17: Cumulative gas production for different F/M ratios (0.5-2) for second set of experiment

F/M ratio	Temperature ºC	Total biogas production (mL)	Net biogas production (mL)	Methane production (mL)	Methane at STP (mL)
2	20	132	78	55	50
1	20	188	66	46	43
0.5	20	223	29	20	19

4.2.2.2 SMA

F/M affects the substrate utilization rate which depends on sludge activity. SMA is estimated by calculating methane production in g CH_4-COD and divided by g VSS added. SMA of different F/M ratios is given in Table 4.18 and 4.19 for both experiments. SMA for F/M ratio 2 is more than other two ratios. This result indicates that F/M ratio of 2 is suitable whereas 0.5 has low SMA. The methane producing capability of sludge at F/M ratio 2 is more as compared to F/M ratios 1 and 0.5. SMA was found to range from 29 to 90 mL CH_4 g^{-1} VSS d^{-1} for F/M varying from 0.5 to 2 for first set of experiment and 32 to 212 mL CH_4 g^{-1} VSS d^{-1} for second set of experiment.

Table 4.18: SMA of different F/M ratios for first set of experiment

F/M ratio	Temperature ºC	Methane (g CH_4 COD)	SMA (g CH_4 COD/g VSS)	SMA (mL CH_4/ g VSS)
2	20	0.081	0.26	90
1	20	0.050	0.11	40
0.5	20	0.051	0.08	29

Table 4.19: SMA of different F/M ratios for second set of experiment

F/M ratio	Temperature ºC	Methane (g CH_4 COD)	SMA (g CH_4 COD/g VSS)	SMA (mL CH_4/ g VSS)
2	20	0.156	0.60	212
1	20	0.132	0.35	123
0.5	20	0.058	0.09	32

CHAPTER 5

CONCLUSIONS

Performance of full scale UASB reactor was evaluated under low temperature conditions at Lalpani, Shimla. Batch test were conducted to evaluate BMP of sewage and SMA of granular sludge. Based on the studies conducted, following conclusions are drawn.

1. The performance of full scale UASB reactor (19.35 MLD) with respect to removal of COD, BOD and Solids under low temperature conditions was not satisfactory as compared to the pilot scale studies reported in the literature under similar temperature conditions. The unsatisfactory performance of sewage treatment plant is attributed to improper operation and maintenance of UASB reactor.
2. Inconsistency in the performance of UASB reactor in terms of removal of suspended solids was due to improper desludging, variations in influent characteristics, incomplete consumption of dissolved organic matter and poor operation and maintenance.
3. In the present study, maximum COD removal efficiency of 42% was obtained in full scale UASB reactor. However, according to the literature, 60-80% of COD reduction has been achieved under psychrophilic conditions when operated with proper operation and maintenance procedures. Similarly, 36% of BOD removal was observed in UASB effluent. Low removal efficiency of the reactor was due to low temperature, which limits the process of hydrolysis subsequently reduces the SMA of sludge resulting in insufficient amount of active settled biomass.
4. Total gas production under controlled conditions (19ºC) for F/M ranging from 0.1 to 4 was less as compared to uncontrolled conditions (0-26ºC). However, higher cumulative methane production was observed in controlled conditions. Frequent fluctuations in temperature affect the methanogenic phase by affecting the methane forming bacteria.
5. BMP of substrate was influenced by F/M ratio. A decreasing trend was noticed in the BMP values when F/M ratio decreased from 4 to 0.35.
6. BMP (F/M ratios 0.1 – 4.0) at constant temperature ($19^{o}C$) was found to be more than BMP at ambient temperature (0 – $26^{o}C$) because the sudden increase (i.e., shock) in temperature leads to decrease in bio-methane producing bacteria.

7. SMA results indicate that 2:1 was the optimal F/M ratio for both batches (SMA-15 day and SMA – 33 day) of experiment because the methane producing capability of sludge at F/M ratio 2 is more as compared to F/M ratios 1 and 0.5.

Recommendations for Further Research Needs

1. Studies should be carried out to enhance the performance of full scale UASB reactor under low temperature conditions treating domestic wastewater.
2. Fecal coliform, Ammonia and Nutrients (nitrogen and phosphorous) removal should be investigated further to improve the effluent quality of UASB reactor under psychrophilic conditions.
3. Studies should be carried out to enhance the granulation of sludge in UASB reactor.

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APPENDIX I

APPENDIX I

A) DATA OF FULL SCALE UASB REACTOR

A1) Sample 1

Date: 23/11/2015

S.NO.	PARAMETERS	UNITS	UASB INLET	UASB OUTLET
1	Temperature of sewage	˚C	18.2	17
2	pH		7.2	7.1
3	BOD_5	mg/l	286	184
4	Alkalinity	mg/l	382	400
5	Dissolved Solids	mg/l	611	668
6	Suspended Solids	mg/l	1863	1150
7	Total Solids	mg/l	2474	1818
8	Volatile Solids	mg/l	584	195
9	COD	mg/l	550	320

A2) Sample 2

Date: 16/12/2015

S.NO.	PARAMETERS	UNITS	UASB INLET	UASB OUTLET
1	Temperature of sewage	˚C	10	11
2	pH		7.28	7.24
3	BOD_5	mg/l	422	318
4	Alkalinity	mg/l	370	462
5	Dissolved Solids	mg/l	617	608
6	Suspended Solids	mg/l	1126	205
7	Total Solids	mg/l	1743	813
8	Volatile Solids	mg/l	1046	360
9	COD	mg/l	736	576

A3) Sample 3

Date: 22/01/2016

S.NO.	PARAMETERS	UNITS	UASB INLET	UASB OUTLET
1	Temperature of sewage	˚C	15	15
2	pH		7.3	7.25
3	BOD_5	mg/l	280	230
4	Alkalinity	mg/l	410	486
5	Dissolved Solids	mg/l	570	605
6	Suspended Solids	mg/l	1009	186
7	Total Solids	mg/l	1579	791
8	Volatile Solids	mg/l	829	511
9	COD	mg/l	536	428

A4) Sample 4

Date: 09/02/2016

S.NO.	PARAMETERS	UNITS	UASB INLET	UASB OUTLET
1	Temperature of sewage	˚C	16	16
2	pH		7.31	7.2
3	BOD_5	mg/l	437	424
4	Alkalinity	mg/l	389	460
5	Dissolved Solids	mg/l	586	607
6	Suspended Solids	mg/l	796	722
7	Total Solids	mg/l	1382	1329
8	Volatile Solids	mg/l	798	734
9	COD	mg/l	768	672

A5) Sample 5

Date: 03/03/2016

S.NO.	PARAMETERS	UNITS	UASB INLET	UASB OUTLET
1	Temperature of sewage	˚C	14	14
2	pH		7.5	7.2
3	BOD_5	mg/l	401	221
4	Alkalinity	mg/l	405	456
5	Dissolved Solids	mg/l	554	603
6	Suspended Solids	mg/l	1009	186
7	Total Solids	mg/l	1563	789
8	Volatile Solids	mg/l	860	288
9	COD	mg/l	576	448

APPENDIX II

APPENDIX II

A) BIOCHEMICAL METHANE POTENTIAL (BMP)

A1) Total Cumulative gas production for F/M ratio 4, 1.3, 0.35 and 0.1 at 19ºC.

Time (days)	F/M Ratio			
	4	1.3	0.35	0.1
0	0	0	0	0
1	0	2.4	5	13
2	10	9.41	23	18
3	10	9.41	23	18
4	19.6	20.16	44.5	35
5	26.6	32.66	58	57
6	32.6	42.66	69.25	72
7	39.1	51.91	80.45	85.6
8	44.2	58.31	88.2	96.6
9	48.8	63.06	94.6	106.7
10	48.8	63.06	94.6	106.7
11	54.8	68.51	102.2	118.7
12	57.8	71.56	106.75	126.2
13	59.8	73.76	110.3	131.7
14	61.8	75.81	113.7	137.1
15	61.8	75.81	116.9	142.3
16	64	78.86	119.85	147.1
17	64	78.86	119.85	147.1
18	66	81.96	124.35	154.2
19	67.1	83.96	127.2	159.2
20	68.1	85.51	129.55	163.2
21	68.1	86.01	130.75	165.6
22	68.1	86.16	131.35	167
23	68.3	86.56	132.35	168.6
24	68.4	87.46	133.6	170.4
25	69.4	88.41	134.85	172.3
26	70.3	89.46	136.15	174.2
27	70.3	89.46	136.15	178

A2) Net Cumulative gas production for F/M ratio 4, 1.3, 0.35 and 0.1 at 19ºC.

Time (days)	F/M Ratio			
	4	**1.3**	**0.35**	**0.1**
0	0	0	0	0
1	0	2.4	-5.3	6.5
2	10	9.41	-9.3	-4.9
3	10	9.41	-9.3	-4.9
4	19.6	17.96	-4.8	-10.9
5	19.6	16.46	-2.3	-18.9
6	23.4	21.46	-0.05	-19.1
7	27.5	25.71	2.65	-19
8	30.4	28.11	5.4	-18.65
9	35	31.86	7.8	-15.15
10	35	31.86	7.8	-15.15
11	40.8	34.31	9.6	-16.1
12	43.2	36.16	11.15	-14.5
13	45.2	37.26	12.5	-13.8
14	47.2	38.71	15.9	-12.6
15	47.2	38.71	19.1	-7.6
16	46.4	39.56	16.25	-11.6
17	46.4	39.56	16.25	-11.6
18	46.4	40.06	15.85	-12.605
19	46.8	41.06	16.6	-11.655
20	47.8	41.81	17.25	-10.255
21	47.8	42.31	18.45	-9.355
22	47.8	42.46	19.05	-8.855
23	48	42.86	18.25	-9.055
24	48.1	42.96	17.9	-8.955
25	49.1	43.21	17.65	-8.805
26	50	43.46	17.55	-8.705
27	50	43.46	17.55	-6.105

A3) Methane production for F/M ratio 4, 1.3, 0.35 and 0.1 at 19ºC.

Time (days)	F/M Ratio			
	4	1.3	0.35	0.1
0	0	0	0	0
1	0	1.68	-3.71	4.55
2	7	6.587	-6.51	-3.43
3	7	6.587	-6.51	-3.43
4	13.72	12.572	-3.36	-7.63
5	13.72	11.522	-1.61	-13.23
6	16.38	15.022	-0.035	-13.37
7	19.25	17.997	1.855	-13.3
8	21.28	19.677	3.78	-13.055
9	24.5	22.302	5.46	-10.605
10	24.5	22.302	5.46	-10.605
11	28.56	24.017	6.72	-11.27
12	30.24	25.312	7.805	-10.15
13	31.64	26.082	8.75	-9.66
14	33.04	27.097	11.13	-8.82
15	33.04	27.097	13.37	-5.32
16	32.48	27.692	11.375	-8.12
17	32.48	27.692	11.375	-8.12
18	32.48	28.042	11.095	-8.8235
19	32.76	28.742	11.62	-8.1585
20	33.46	29.267	12.075	-7.1785
21	33.46	29.617	12.915	-6.5485
22	33.46	29.722	13.335	-6.1985
23	33.6	30.002	12.775	-6.3385
24	33.67	30.072	12.53	-6.2685
25	34.37	30.247	12.355	-6.1635
26	35	30.422	12.285	-6.0935
27	35	30.422	12.285	-4.2735

A4) Total Cumulative gas production for F/M ratio 4, 1.3, 0.35 and 0.1 at 0-26ºC.

Time (days)	F/M Ratio			
	4	**1.3**	**0.35**	**0.1**
0	0	0	0	0
1	1.325	2.6	6.3	4.8
2	2.825	5.73	10	13.7
3	4.925	9.2	16.1	23.7
4	4.925	9.2	16.1	23.7
5	9.325	15.6	26.1	38
6	11.725	19.4	32.65	48.3
7	14.625	23.8	39.55	60.5
8	17.575	28.2	46.65	72.6
9	21.075	33.2	54.85	85.3
10	24.175	38.13	62.55	96.9
11	24.175	38.13	62.55	96.9
12	29.675	45.7	74.35	114.2
13	33.475	51.16	84.05	130.8
14	35.875	54.9	90.6	140.9
15	38.475	58.96	97.5	151.8
16	41.375	63.36	105.1	163.5
17	43.375	66.63	110.3	173.5
18	43.375	66.63	110.3	173.5
19	47.675	72.76	121	190.2
20	50.475	77.63	129.1	202.5
21	53.375	82.03	136.7	214.5
22	55.975	85.63	143.7	225.8
23	59.475	90.83	152.4	239.4
24	61.875	94.83	159.4	250.3
25	61.875	94.83	159.4	250.3
26	64.475	99.56	168.15	263.6
27	66.475	102.96	173.85	273.3
28	68.675	106.2	180.15	283.1
29	70.775	109.53	186.25	292.8
30	72.875	112.93	192.45	303

31	72.875	112.93	198.35	312.6

A5) Net Cumulative gas production for F/M ratio 4, 1.3, 0.35 and 0.1 at 0-26ºC.

Time (days)	F/M Ratio			
	4	1.3	0.35	0.1
0	0	0	0	0
1	1.325	-2.2	-3.7	-8.6
2	2.825	0.93	-4.4	-11.5
3	4.925	4.4	-2.1	-13.1
4	4.925	4.4	-2.1	-13.1
5	9.325	8.6	-2.1	-17.8
6	11.725	10.3	-1.55	-20.9
7	14.425	9.9	-1.25	-22.1
8	14.775	10.7	-0.95	-22.4
9	15.075	11.9	-1.55	-26.7
10	15.775	14.03	-1.05	-29
11	15.775	14.03	-1.05	-29
12	17.675	12.4	-2.05	-35.1
13	18.275	13.06	-0.55	-33.5
14	20.675	15.6	2	-32.6
15	21.675	16.66	2.8	-32.5
16	24.575	17.46	4.2	-33.6
17	24.975	18.73	6.2	-33.6
18	24.975	18.73	6.2	-33.6
19	24.675	18.46	5.5	-37.3
20	25.875	19.73	8.4	-37.8
21	25.975	20.13	7.8	-38.8
22	27.175	20.73	8.8	-34.5
23	27.075	21.13	8.3	-35.9
24	28.475	22.73	10.1	-38
25	28.475	22.73	10.1	-38
26	29.275	24.06	9.85	-42.1
27	31.275	25.06	10.35	-42.4
28	29.675	25.5	11.25	-41.2

29	30.375	26.23	12.35	-42.9
30	30.775	26.63	13.55	-44.5
31	30.775	26.63	13.65	-46.5

A6) Methane production for F/M ratio 4, 1.3, 0.35 and 0.1 at 0-26ºC.

Time (days)	F/M Ratio			
	4	**1.3**	**0.35**	**0.1**
0	0	0	0	0
1	0.93	-1.54	-2.59	-6.02
2	1.98	0.653	-3.08	-8.05
3	3.45	3.08	-1.47	-9.17
4	3.45	3.08	-1.47	-9.17
5	6.53	6.02	-1.47	-12.46
6	8.21	7.21	-1.085	-14.63
7	10.1	6.93	-0.875	-15.47
8	10.3	7.49	-0.665	-15.68
9	10.6	8.33	-1.085	-18.69
10	11	9.823	-0.735	-20.3
11	11	9.823	-0.735	-20.3
12	12.4	8.68	-1.435	-24.57
13	12.8	9.147	-0.385	-23.45
14	14.5	10.92	1.4	-22.82
15	15.2	11.67	1.96	-22.75
16	17.2	12.23	2.94	-23.52
17	17.5	13.11	4.34	-23.52
18	17.5	13.11	4.34	-23.52
19	17.3	12.93	3.85	-26.11
20	18.1	13.81	5.88	-26.46
21	18.2	14.09	5.46	-27.16
22	19	14.51	6.16	-24.15
23	19	14.79	5.81	-25.13
24	19.9	15.91	7.07	-26.6
25	19.9	15.91	7.07	-26.6
26	20.5	16.85	6.895	-29.47

27	21.9	17.55	7.245	-29.68
28	20.8	17.85	7.875	-28.84
29	21.3	18.36	8.645	-30.03
30	21.5	18.64	9.485	-31.15
31	21.5	18.64	9.555	-32.55

B) SPECIFIC METHANOGENIC ACTIVITY (SMA-1)

B1) Total Cumulative gas production for F/M ratio 2, 1 and 0.5 at 20ºC.

Time (days)	F/M Ratio		
	2	1	0.5
0	0	0	0
1	8.1	4	1.47
2	22.55	11.87	4.37
3	22.55	11.87	4.37
4	50.65	25.57	15.5
5	68.95	37.97	22.1
6	85.7	50.47	28.7
7	97.7	60.93	34.4
8	107.4	68	37.5
9	116.2	75.2	41.5
10	116.2	75.2	41.5
11	127.3	86.6	49
12	137	96.33	56.1
13	144.9	105.5	63.7
14	151.8	113.4	71.6
15	151.8	115.5	79.9

B2) Net Cumulative gas production for F/M ratio 2, 1and 0.5 at 20ºC.

Time (days)	F/M Ratio		
	2	1	0.5
0	0	0	0
1	-6.15	-19.55	-5.33
2	-7.3	-19.18	-2.43
3	-7.3	-19.18	-2.43
4	-8.95	-24.88	1.467
5	-3.65	-20.88	3.9
6	4.9	-14.93	6.733
7	8.6	-6.467	10.4
8	13.8	0.3	12.33
9	17.9	3.2	14.07
10	17.9	3.2	14.07
11	22.2	10.2	19.6
12	24.3	14.933	25.87
13	26.1	20.367	29.07
14	27.2	24.767	35
15	27.2	26.9	43.33

B3) Methane production for F/M ratio 2, 1and 0.5 at 20ºC.

Time (days)	F/M Ratio		
	2	**1**	**0.5**
0	0	0	0
1	-4.305	-13.68	-3.73
2	-5.11	-13.42	-1.70
3	-5.11	-13.42	-1.70
4	-6.265	-17.41	1.02
5	-2.555	-14.61	2.73
6	3.43	-10.45	4.71
7	6.02	-4.52	7.28
8	9.66	0.21	8.63
9	12.53	2.24	9.84
10	12.53	2.24	9.84
11	15.54	7.14	13.72
12	17.01	10.45	18.10
13	18.27	14.25	20.34667
14	19.04	17.33	24.5
15	19.04	18.83	30.33333

C) SPECIFIC METHANOGENIC ACTIVITY (SMA-2)

C1) Total Cumulative gas production for F/M ratio 2, 1 and 0.5 at 20ºC.

Time (days)	F/M Ratio		
	2	**1**	**0.5**
0	0	0	0
1	6.4	3.7	1.2
2	16.4	8.7	3.2
3	16.4	8.7	3.2
4	34.3	21	9.5
5	47.4	29.6	13.5
6	60.1	39.4	18
7	72.4	49.6	22.9
8	83.4	60.7	28.8
9	94.3	71.2	35
10	94.3	71.2	35
11	107.3	88.5	47.2
12	117.5	101.4	55.9
13	125.25	109.95	62.7
14	132.85	117.25	70.2

15	141.65	124.85	79
16	149.05	130.55	86.1
17	149.05	130.55	86.1
18	161.45	139.55	97.3
19	168.85	144.65	103.4
20	175.15	148.95	108.1
21	180.35	152.85	111.4
22	186.55	157.85	115.3
23	191.65	161.5	117.5
24	191.65	161.5	117.5
25	198.85	167.35	120.7
26	204.55	171.75	123.3
27	209.15	175.85	125.2
28	212.35	178.15	126.2
29	217.05	181.55	128
30	221.55	183.25	130.1
31	221.55	184.45	130.1
32	224.75	186.05	131.1
33	224.75	187.65	132.1

C2) Net Cumulative gas production for F/M ratio 2, 1 and 0.5 at 20ºC.

Time (days)	**F/M Ratio**		
	2	**1**	**0.5**
0	0	0	0
1	-4.7	-3.1	-1.8
2	0.5	1.9	0.2
3	0.5	1.9	0.2
4	0.5	2.6	6.5
5	4.1	6.2	9.3
6	8	9.6	11.3
7	11.5	13.4	13
8	12.3	17.9	14.95
9	14.4	22.2	17.9
10	14.4	22.2	17.9
11	10.6	28.3	23.5
12	9.4	34.4	28.5
13	10.55	39.75	33.8
14	10.55	42.05	39.3

15	10.65	43.45	45
16	12.05	45.55	50.3
17	12.05	45.55	50.3
18	12.45	47.15	57.9
19	15.2	49.45	63.5
20	16.7	50.55	67.4
21	18.6	52.45	69.1
22	18.6	52.65	69.6
23	20.9	54.7	71.8
24	20.9	54.7	71.8
25	21.4	56.95	74.4
26	22.6	58.55	74.4
27	23.9	60.25	75.3
28	26.2	62.55	75.6
29	26.8	62.95	76.2
30	26.9	61.45	76.05
31	26.9	62.65	76.05
32	29.1	64.25	77.05
33	29.1	65.85	78.05

C3) Cumulative Methane production for F/M ratio 2, 1 and 0.5 at 20°C.

Time (day)	F/M Ratio		
	2	1	0.5
0	0	0	0
1	-3.29	-2.17	-1.26
2	0.35	1.33	0.14
3	0.35	1.33	0.14
4	0.35	1.82	4.55
5	2.87	4.34	6.51
6	5.6	6.72	7.91
7	8.05	9.38	9.1
8	8.61	12.53	10.465
9	10.08	15.54	12.53
10	10.08	15.54	12.53
11	7.42	19.81	16.45
12	6.58	24.08	19.95
13	7.385	27.825	23.66
14	7.385	29.435	27.51

15	7.455	30.415	31.5
16	8.435	31.885	35.21
17	8.435	31.885	35.21
18	8.715	33.005	40.53
19	10.64	34.615	44.45
20	11.69	35.385	47.18
21	13.02	36.715	48.37
22	13.02	36.855	48.72
23	14.63	38.29	50.26
24	14.63	38.29	50.26
25	14.98	39.865	52.08
26	15.82	40.985	52.08
27	16.73	42.175	52.71
28	18.34	43.785	52.92
29	18.76	44.065	53.34
30	18.83	43.015	53.235
31	18.83	43.855	53.235
32	20.37	44.975	53.935
33	20.37	46.095	54.635