Asian Journal of Water, Environment and Pollution, Vol. 11, No. 2 (2014), pp. 67–79.

Performance Studies on Biological Treatment of Slaughterhouse Wastewater Using Mixed Culture in Sequencing Batch Reactor

Pradyut Kundu, Anupam Debsarkar[1] and Somnath Mukherjee[1*]
A.P.C. Roy Polytechnic, Jadavpur, Kolkata – 700 032
[1]Civil Engineering Department, Jadavpur University, Kolkata – 700 032
✉ snm_ju@yahoo.co.in

Received April 4, 2013; revised and accepted February 22, 2014

Abstract: The slaughterhouse and meat processing units generate large volumes of wastewater containing high organic and nitrogenous substances (COD, NH_4^+-N), which require considerable degree of treatment before discharge to the water body. In this regard, selecting an effective treatment system is important. Amongst various biological treatment systems sequencing batch reactor (SBR) is comparatively noble bioreactor system for treating waste containing carbon and nitrogen simultaneously. The performance of a 20 L sequencing batch reactor (SBR) treating wastewater discharged from a local small-scale slaughterhouse was examined in the laboratory at ambient temperature. The reactor was operated under three different variations of aerobic-anoxic sequence, viz. 4 + 4, 5 + 3 and 3 + 5 hours of total react period with influent soluble COD (SCOD) and ammonia nitrogen level 1000 ± 50 mg/L & 90 ± 10 mg/L and 2000 ± 50 mg/L & 180 ± 10 mg/L, respectively. It has been observed that 80 to 96% of SCOD removal would be possible at the end of eight hours of overall reaction period, irrespective of the length of the aerobic react period. In case of 4+4 aerobic-anoxic operating cycle, reasonable degree of nitrification 89.48% and 81.58% corresponding to initial NH_4^+-N value of 87.52 mg/L and 185.24 mg/L respectively, along with 94.07% and 90.23% of organic carbon removal corresponding to initial SCOD value of 1015.24 mg/L and 2028.55 mg/L respectively, have been achieved after eight hours of react period for treatment of slaughterhouse wastewater in SBR.

Key words: Slaughterhouse wastewater, sequencing batch reactor, carbon oxidation, ammonia oxidation.

Introduction

The slaughterhouse and meat processing industry generates large volumes of wastewater containing diluted blood, protein, fat and suspended solids which require considerable degree of treatment before getting released to the water environment. Organic carbon and nitrogen are major contaminants of concern in the wastewater. Slaughterhouse wastewater contains high-strength organic carbon (C), nitrogen (N) and phosphorus (P). After preliminary and primary treatment, it contains suspended solids (SS) of 400–2000 mg/L, chemical oxygen demand (COD) of 1000–8000 mg/L, total nitrogen (TN) of 150–400 mg/L, total phosphorus (TP) of 20–80 mg/L, and fats of 20–300 mg/L (Cassidy and Belia, 2005; Del Pozo and Diez, 2005; Mittal, 2006).

In India, with the advent of economical growth of industrial waste, pollution attributes a significant problem to environmental engineers and scientists. Conventional treatment practice applying activated sludge process (ASP) may effectively treat both municipal and industrial wastewater, but their high cost owing to large land requirements often make them economically unattractive. Under such circumstances,

*Corresponding Author

sequencing batch reactor (SBR) may stand as a good option for becoming a viable alternative and thus research on SBR is very much needed in Indian context, particularly to treat both COD and ammonia nitrogen-contaminated wastewater.

SBR technology is a periodic fill and draw type activated sludge process. In SBR system the reaction tank is filled with wastewater for a discrete period of time. The liquid is then allowed to undergo biochemical reaction in the reactor for a definite period of time in a batch mode of operation under aerobic and anoxic condition, which is known to be a react time in presence of active microorganism. Then the mixed liquor is allowed to settle after the react period and the clarified effluent is withdrawn after a time period known as settle period. A properly designed SBR can simulate any conventional continuous flow activated sludge system by providing multiple SBR tanks in parallel. Each SBR responses the functions of equalization, aeration and sedimentation in the same vessel. In continuous flow systems the above practices are conventionally done in the separate tanks. Each tank in SBR system undergoes one or more cycles (i.e. the time between one filling and the next succession) during each day. In the present study, an attempt has been made to explore the performance efficacy of SBR technology for simultaneous removal of soluble carbonaceous organic matter and ammonia nitrogen from slaughterhouse wastewater.

Sequencing Batch Reactor Process Cycles

The operating principles of a sequencing batch reactor (SBR) are characterized in five discrete periods shown in Figure 1.

Fill Period: The influent wastewater is distributed throughout the settled sludge through the influent distribution manifold to provide good contact between the microorganisms and the substrate. The influent can be either pumped in or allowed to flow in by gravity. Most of this period occurs without aeration to create an environment that favours the procreation of microorganisms with good settling characteristics.

React Period: During this period aeration continues until complete biodegradation of organic carbon and nitrogen is achieved. After the substrate is consumed famine stage starts. During this stage some microorganisms will die because of the lack of food and will help reduce the volume of the settling sludge.

Settle Period: Aeration and mixing are discontinued at this stage and solids separation takes place leaving clear, treated effluent above the sludge blanket. During this clarifying period no liquids should enter or leave the tank to avoid turbulence in the supernatant.

Decant Period: This period is characterized by the withdrawal of treated effluent from approximately two feet below the surface of the mixed liquor by the floating solids excluding decanter. This removal must be done without disturbing the settled sludge.

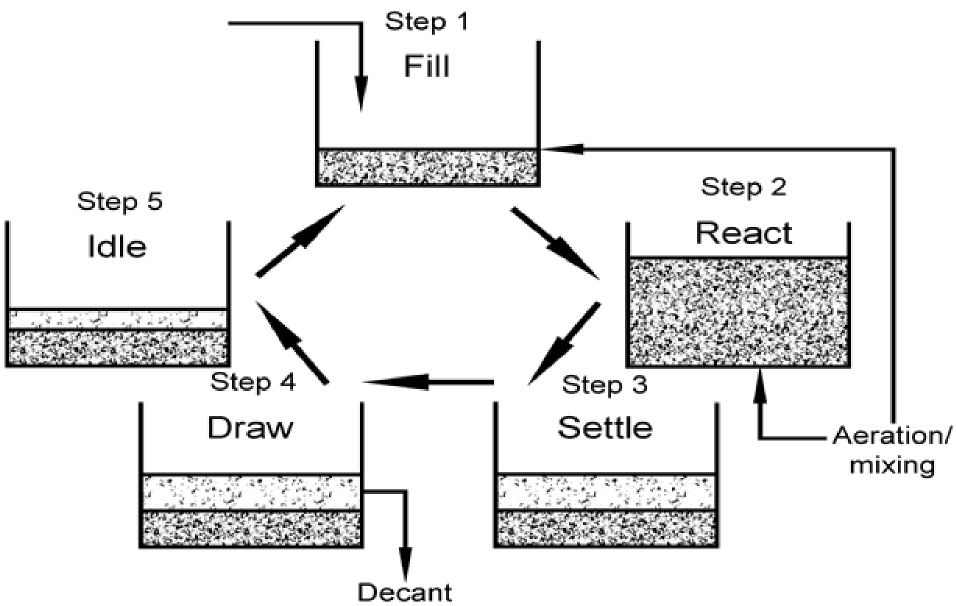

Figure 1: SBR operation cycle showing five discrete periods as Fill, React, Settle, Draw and Idle.

Idle Period: The time in this stage can be used to waste sludge or perform backwashing of the jet aerator. The wasted sludge is pumped to an anaerobic digester to reduce the volume of the sludge to be discarded. The frequency of sludge wasting ranges between once every two or three months depending upon system design.

Advantages of SBR

SBR possesses following advantages over conventional biological treatment units.

(i) It serves as an equalizing basin during fill period and can tolerate waste shock loads under extreme conditions.
(ii) Recycling of sludge is not necessary.
(iii) Solid-liquid separation occurs under nearly ideal quiescent conditions. Hence short circuiting is non-existing during settling.
(iv) Changing the fill level easily controls filamentous growth.
(v) SBR can be operated to achieve nitrification/denitrification or phosphorous removal without chemical addition in the same reactor.

Biological treatment of wastewater containing organic carbon and nitrogen (COD and TKN) have been carried out in laboratory and pilot scale experiment by several researchers successfully (Al-Mutairi et al., 2007; Boopathy et al., 2007; Kim et al., 2008; Al-Mutairi et al., 2008; Lemaire et al., 2009; Roy et al., 2010; Rajagopol et al., 2011; Palatsi et al., 2011; Kern et al., 2012; Wang et al., 2012). According to Kargi and Uygur (2002), biological treatment is the best alternative to treat high strength wastewater containing both carbonaceous organic matter and nutrients (COD and TKN). Mahvi et al. (2004) carried out a pilot-scale study on removal of nitrogen both from synthetic and domestic wastewater in a continuous flow SBR and obtained a total nitrogen and TKN removal of 70-80% and 85-95%, respectively. The performance of a 10 L sequencing batch reactor (SBR) treating wastewater discharged from a local slaughterhouse was examined by Li et al. (2006) in the laboratory at ambient temperature. The influent wastewater contained 3360 mg/L of COD, 190 mg/L of total nitrogen (TN) and 22 mg/L of total phosphorus (TP) on average basis and they achieved COD, TN and TP removal efficiencies of 98, 92 and 98%, respectively. The treatment of slaughterhouse wastewater with co-coagulation, air-flotation and SBR process was studied by Wang Xiao et al. (2009). After co-coagulation flotation process, COD and NH_4^+-N concentrations of 2500 and 160 mg/L of the influent were reduced 90 and 60%, respectively and the biodegradability of wastewater was improved.

The objective of the present work is to conduct the performance study of laboratory-scale sequencing batch reactor (SBR) for combined removal of soluble organics (COD) and nitrogen from slaughterhouse wastewater by changing operational conditions like MLVSS concentration, initial substrate concentration, DO level, pH level, anoxic/aerobic sequence, anoxic/aerobic time phase, etc.

Biological Treatment of Organic Waste and Nitrogen

Biological Treatment of Organic Waste

The removal of carbonaceous COD includes the coagulation of non-settleable colloidal solids and stabilization of organic matter. The latter one is accomplished by microorganisms, principally by chemo heterotrophic carbonaceous bacteria. The microorganisms convert the colloidal and dissolved carbonaceous organic matter into simple end products and additional biomass.

$$v_1 \text{ (organic)} + v_2O_2 + v_3NH_3 + v_4PO_4^{3-} + \text{microorganisms} \longrightarrow v_5 \text{ (new cells)} + v_6CO_2 + v_7H_2O$$

where v_i is stoichiometric coefficient.

Biological Treatment of Nitrogen

Biological nitrogen removal can be achieved by three major biological processes: ammonification, nitrification, and denitrification.

Ammonification: Ammonification occurs when organic nitrogen is converted to ammonia. It is an important mechanism that ultimately allows organic nitrogen to be removed from wastewaters through hydrolysis to amino acids, which are broken down to produce ammonium or directly incorporated into biosynthetic pathways in support of bacterial growth.

Nitrification: Nitrification is the two-step process for the conversion of ammonium nitrogen in the form of ammonia or ammonium to the form of nitrate or nitrite which is shown in Figure 2. Ammonium nitrogen is oxidized to nitrite by ammonia oxidizing bacteria (AOB) and then to nitrate by nitrite oxidizing bacteria (NOB). Many AOBs and NOBs are autotrophic, although heterotrophic bacteria are known to function as nitrifiers (Painter, 1977). The principal genera of importance in biological nitrification processes are *Nitrosomonas, Nitrobacter, Nitrosospira, Nitrosolobus, Nitrosovibrio*, and *Nitrosococcus* (Madigan et al., 2000). These genera of organisms are autotrophic, so their carbon source is carbon dioxide (CO_2).

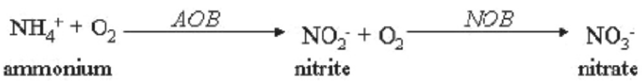

Figure 2: Biological transformation of ammonium nitrogen.

Denitrification: Denitrification involves conversion of nitrate or nitrite into various reduced nitrogen compounds, mostly in the form of nitrogen gas. Microorganisms utilize the nitrate as an electron acceptor in the absence of oxygen. The reactions for denitrifying bacteria with glucose as carbon source are given by:

$$C_6H_{12}O_6 + 12NO_3^- \longrightarrow 6CO_2 + 12NO_2^- + 6H_2O + \text{energy}$$

$$C_6H_{12}O_6 + 8\ NO_2^- + 8H^+ \longrightarrow 6CO_2 + 4N_2 + 10H_2O + \text{energy}$$

Many different species of bacteria have the capability of denitrification, including those from *Pseudomonas, Achromobacter, Bacillus, Rhizobium, Aquaspirillum, Flavobacterium* and *Aeromonas.*

Materials and Methods

Seed Acclimatization for Combined Carbon Oxidation, Nitrification and Denitrification

Seed acclimatization study was conducted in a measuring cylinder of 2.0 L volume. Heterotrophic microbes were actively acclimatized in laboratory environment by inoculating sludge collected from an aeration pond of Mokami small-scale slaughterhouse located in the village Nazira, South 24 Parganas district (West Bengal) India, to a growth propagating media composed of 250 ml of nutrient solution. The composition of the nutrient solution in 1000 ml distilled water comprised 60.0 mg K_2HPO_4, 40.0 mg KH_2PO_4, 500.0 mg $MgSO_4.7H_2O$, 710.0 mg $FeCl_3.6H_2O$, 0.1 mg $ZnSO_4.7H_2O$, 0.1 mg $CuSO_4.5H_2O$, 8.0 mg $MnCl_2.2H_2O$, 0.11 mg $(NH_4)_6Mo_7O_{24}$, 100.0 mg $CaCl_2.2H_2O$, 200.0 mg $CoCl_2.6H_2O$, 55.0 mg $Al_2(SO_4)_3.16H_2O$ and 150.0 mg H_3BO_3. Finally 750 ml volume of distilled water was added to liquid mixture to make a volume of 1 L and the mixture was continuously aerated with intermittent feeding with dextrose solution having concentrations of 1000 mg/L and ammonium chloride (NH_4Cl) having concentration of 100 mg/L as a carbon and nitrogen source respectively. The acclimatization process was continued for an overall period of 60 days. The biomass growth was indicated by the magnitude of sludge volume index (SVI) and mixed liquor volatile suspended solid (MLVSS) concentration in the reactor. pH in the reactor was maintained in the range 6.8-7.5 by adding required amount of sodium carbonate (Na_2CO_3) and phosphate buffer. The seed acclimatization phase was considered to be over when a steady state condition were observed in terms of equilibrium COD and NH_4^+-N reduction with respect to a steady level of MLVSS concentration in the reactor.

Denitrifying seed was cultured separately in 2.0 L aspirator bottle. 500 gm of digested sludge obtained from a local sewage treatment plant (STP) was added to 1.0 L of distilled water. The solution was filtered and added to 500 ml of untreated sewage. The resulting solution was acclimatized for denitrification purpose using dextrose as carbon source having concentrations of 1000 mg/L and potassium nitrate (KNO_3) as the source of nitrate-nitrogen (NO_3^--N) having concentrations of 20 mg/L. A magnetic stirrer was provided for proper mixing of the solution. Denitrifying seed was acclimatized against a nitrate-nitrogen concentration varying from 20-50 mg/L as N, over a period of two months.

Experimental Setup

The experimental work was carried out in a laboratory scale SBR, made of Perspex sheet of 6 mm thickness, having 20 L of effective volume. The feed solution is kept in a 25 L capacity carbuoy placed on an elevated platform of wooden type. A feed tube as inlet is connected from bottom of reservoir to inlet spout of reactor. Oxygen was supplied through belt driven small air compressor. A stirrer of 0.3 kW capacity was installed at the centre of the vessel for mixing the content of the reactor. Air supply was provided during aerobic phase of react period. However, during the anoxic phase the stirrer was allowed to operate for mixing purpose. A timer was also connected to compressor for controlling the sequence of different react period (aerobic and anoxic). Oxygen was supplied in the reactor through strainer type diffuser, placed at the bottom of the reactor. A schematic diagram of the experimental set-up is shown in Figure 3.

Experimental Procedure for Combined Carbon Oxidation, Nitrification and Denitrification

In the present study 750 ml of previously acclimatized combined carbonaceous and nitrogenous seed along with 250 ml of previously acclimatized denitrifying seed were added to slaughterhouse wastewater sample

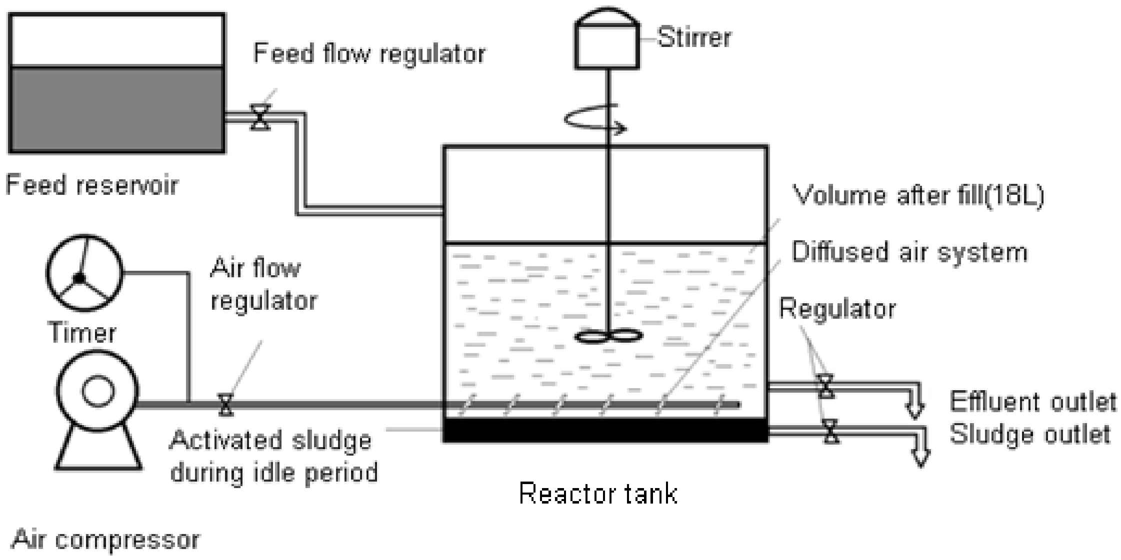

Figure 3: Schematic diagram of the experimental setup.

in a sequential batch reactor of 20.0 L effective capacity having initial soluble COD (SCOD) and ammonia-nitrogen level (NH_4^+-N) concentration in the range of level 1000 ± 50 mg/L & 90 ± 10 mg/L and 2000 ± 50 mg/L & 180 ± 10 mg/L, respectively. At the beginning of the experiment, pH in the reactor was maintained in the range of 6.8-7.5 by adding required amount of sodium carbonate (Na_2CO_3) and phosphate buffer. The cycle period for the operation of SBR was taken as 10 hours, with a fill period of 0.5 hour, overall react period of eight hours, settle period of one hour and idle/decant period 0.5 hour. The overall react period was divided into aerobic and anoxic react period in the following sequences.

Combination 1: 4-hour aerobic react period and 4-hour anoxic react period.

Combination 2: 5-hour aerobic react period and 3-hour anoxic react period.

Combination 3: 3-hour aerobic react period and 5-hour anoxic react period.

During the time course of the study, 100 ml of sample was collected from the outlet of the reactor at every one hour interval, on completion of the fill period. The samples were analyzed for the following parameters, viz. pH, DO, MLSS, MLVSS, COD, NH_4^+-N, NO_2^--N and NO_3^--N as per the methods described in Standard Methods (APHA, 1991). The pH of the solution was measured by a digital pH meter (Systronics, India make). NH_4^+-N and NO_3^--N were estimated by respective ion selective electrodes in Orion ISE meter. COD was analyzed by closed reflux method using dichromate digestion principle and HACH, USA digester. Dissolved oxygen (DO) was measured electrometrically by digital DO meter (Systronics, India make). Mixed liquor suspended solids (MLSS) and mixed liquor volatile suspended solids (MLVSS) were measured by gravimetric method at temperature of 103-105 °C and 550 ± 50 °C in muffle furnace, respectively.

Results and Discussions

Characterization of Slaughterhouse Wastewater

The slaughterhouse wastewater samples were collected from two different locations: (i) the raw (untreated) wastewater from the main collection pit and (ii) the primary treated effluent from the inlet box of aeration basin. The wastewater samples were collected five times over the entire course of the study in 20 L plastic containers and stored in a refrigerator at approximately 4.0 ºC. The above wastewater samples (raw and primary treated) were characterized in the laboratory with respect to the parameters as exhibited in Tables 1 and 2 respectively.

The performance of the present study was carried out with wastewater having an average initial soluble COD (SCOD) and ammonia-nitrogen (NH_4^+-N) concentration of 1000 ± 50 mg/L & 90 ± 10 mg/L and 2000 ± 50 mg/L & 180 ± 10 mg/L respectively.

Carbon Oxidation Profile of Slaughterhouse Wastewater in SBR

Organic carbon, which is the source of energy for microbial metabolism, has been estimated in terms of chemical oxygen demand (COD). In the case of a

Table 1: Characteristics and composition of raw slaughterhouse wastewater

Parameters	*Set 1*	*Set 2*	*Set 3*	*Set 4*	*Set 5*	*Mean*	*Standard deviation*
pH	8.0	7.5	7.8	7.6	8.5	7.88	0.3544
Total Suspended Solids (TSS)	10120	11225	12548	10357	14225	11695	1524.48
Total Dissolved Solids (TDS)	6345	6687	7152	6875	7840	6979.80	503.84
BOD_5 at 20 °C	3000	3150	3200	3420	3500	3254	182.38
SCOD	5185	5540	5860	6428	6840	5970.60	596.63
Total Kjeldahl Nitrogen (TKN)	850	910	980	1054	1150	988.80	105.63
NH_4^+-N (as N)	450	500	540	610	650	550	72.38

All units are in mg/L except pH.

Table 2: Characteristics and composition of primary treated slaughterhouse wastewater

Parameters	*Set 1*	*Set 2*	*Set 3*	*Set 4*	*Set 5*	*Mean*	*Standard deviation*
pH	7.5	7.8	8.0	7.6	8.5	7.88	0.35
Total Suspended Solids (TSS)	4255	4550	4842	5075	5340	4812.40	381.50
Total Dissolved Solids (TDS)	3800	3880	3958	4125	4230	3998.60	157.91
BOD_5 at 20 °C	710	950	1180	1420	1565	1165	309.35
SCOD	1030	1250	1580	1870	2056	1557.20	379.33
Total Kjeldahl Nitrogen (TKN)	315	350	410	445	485	401	61.75
NH_4^+-N (as N)	92	110	128	155	188	134.60	33.85

All units are in mg/L except pH.

particular experiment, carried out with an initial SCOD of 1015.24 mg/L and initial NH_4^+-N concentration of 87.52 mg/L, it has been observed that the major fraction of SCOD removal (60.09%) took place during aerobic react period of four hours. In the anoxic phase, further SCOD removal has been noticed to the extent of 94.07% (Figure 4). A similar trend of removal pattern was also observed in Figure 5 when initial NH_4^+-N concentration was increased to 185.24 mg/L as N with initial SCOD of 2028.55 mg/L in a separate set of experiment. It is revealed from Figures 4 and 5 that SCOD level has decreased rapidly during aerobic react period as compared to its rate of removal during anoxic condition due to dominance of organic heterotrophs. When the react period was changed into 5-hour aerobic followed by a reduced 3-hour anoxic, a marginal improvement of SCOD removal in aerobic phase (78.95%) and anoxic phase (96.07%) with an initial SCOD of 1023.22 mg/L was observed due to enhanced aeration time. On the other hand, when the react period was subsequently changed to 3-hour aerobic period followed by 5-hour anoxic period, a marginal decrease of SCOD removal in aerobic phase (59.85%) and anoxic phase (81.26%) with an initial SCOD of 1042.52 mg/L was obtained due to lag of aeration time.

Ammonia Oxidation Profile of Slaughterhouse Wastewater in SBR

Ammonia oxidation took place due to the presence of previously acclimatized nitrifying organisms within the reactor. Considering the cycle period of four hours (aerobic) and four hours (anoxic), it has been observed that at the end of 8-hour react period of reaction, ammonia oxidation was 89.48 and 81.58%, when initial NH_4^+-N was approximately 87.52 and 185.24 mg/L as N respectively. The ammonia oxidation occurred in two phases. A fraction of ammonia was assimilated by cell-mass for synthesis of new cell during carbon oxidation. The dissimilatory removal of ammonia depends on the population of nitrifiers and oxidation time. The descending trend of ammonia removal for higher level of initial concentration of NH_4^+-N was mainly due to limitation of enzymatic metabolism of nitrifiers. When the reactor system was operated in 5-hour (aerobic) and 3-hour (anoxic) mode of react cycle, an overall improvement of ammonia oxidation from 89.48 to 95.94 and 83.52% for initial NH_4^+-N of 93.54 and 173.88 mg/L as N respectively, was observed. The results reveal the fact that the extension of aeration period helped to enhance the oxidation efficiency for the present system.

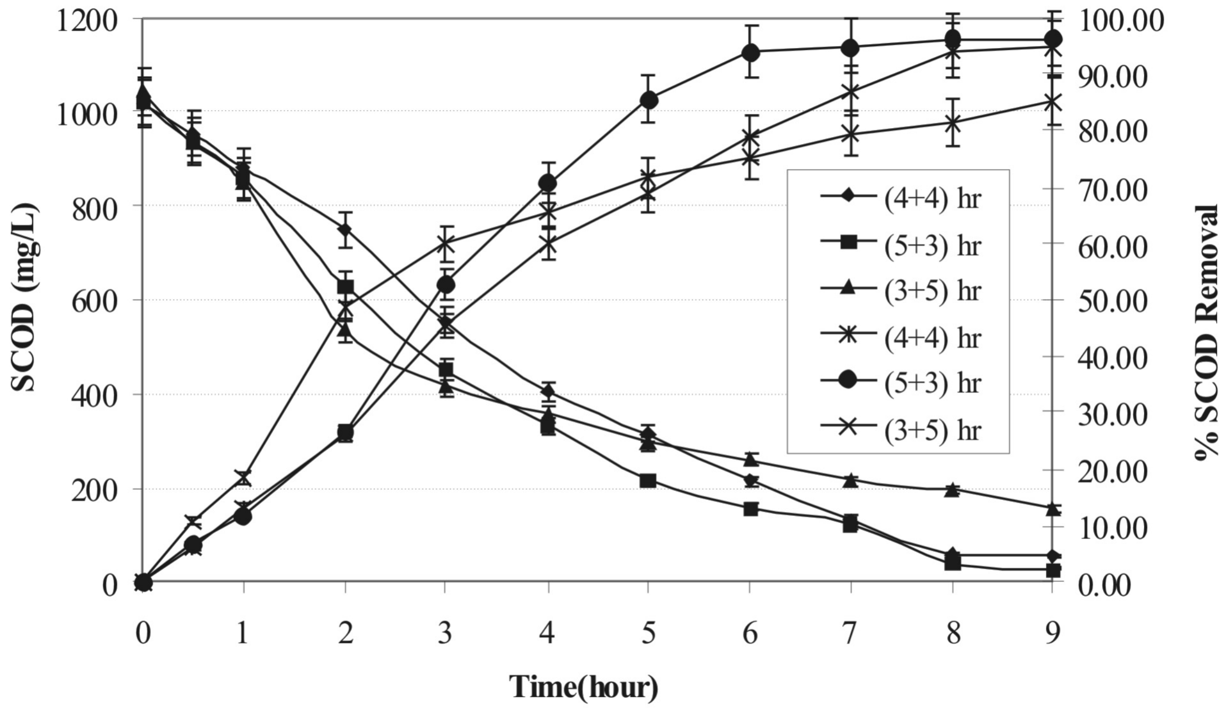

Figure 4: Carbon oxidation profiles in SBR under different react period combination [initial SCOD = 1000 ± 50 mg/L and initial NH_4^+-N = 90 ± 10 mg/L as N].

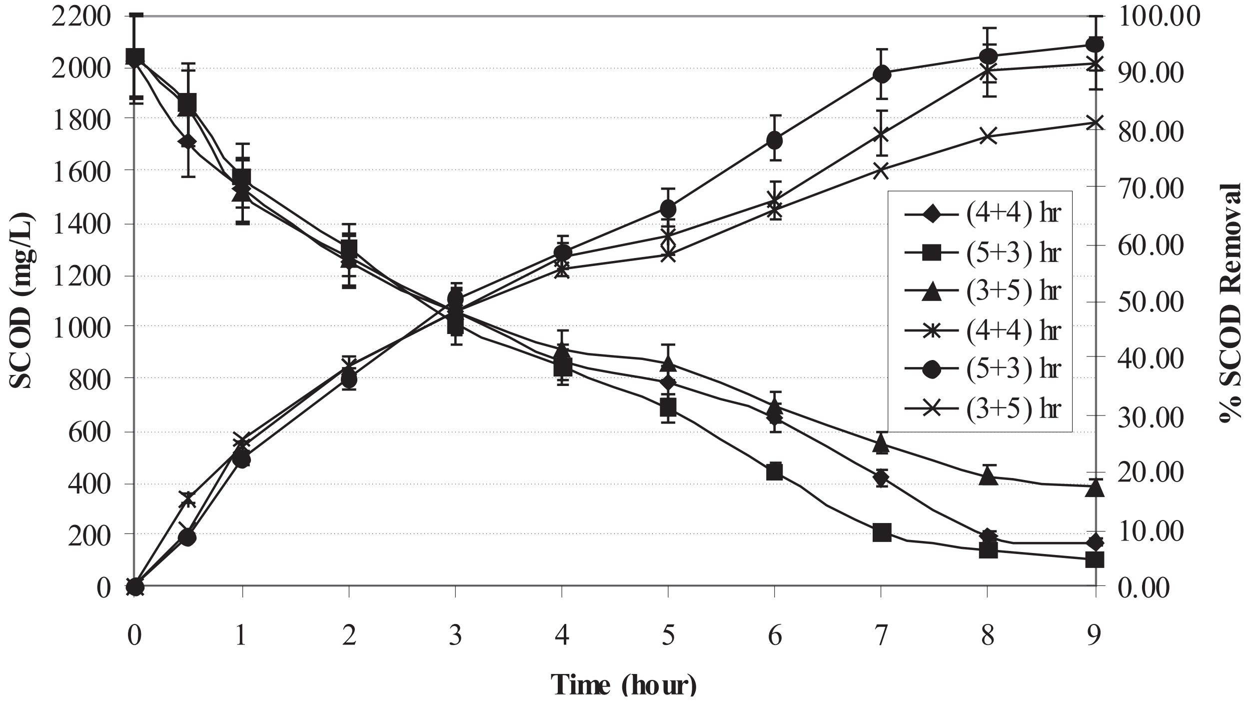

Figure 5: Carbon oxidation profiles in SBR under different react period combination [initial SCOD = 2000 ± 50 mg/L and initial NH_4^+-N = 180 ± 10 mg/L as N].

It was also observed that when aerobic period was reduced to three hours, ammonia oxidation reduced to 77.67 and 68.21% corresponding to initial NH_4^+-N value of 96.58 and 176.85 mg/L as N respectively, at the end of 8-hour react period. The NH_4^+-N level at different time periods for different combinations of react periods and initial NH_4^+-N concentration are exhibited in Figures 6 and 7.

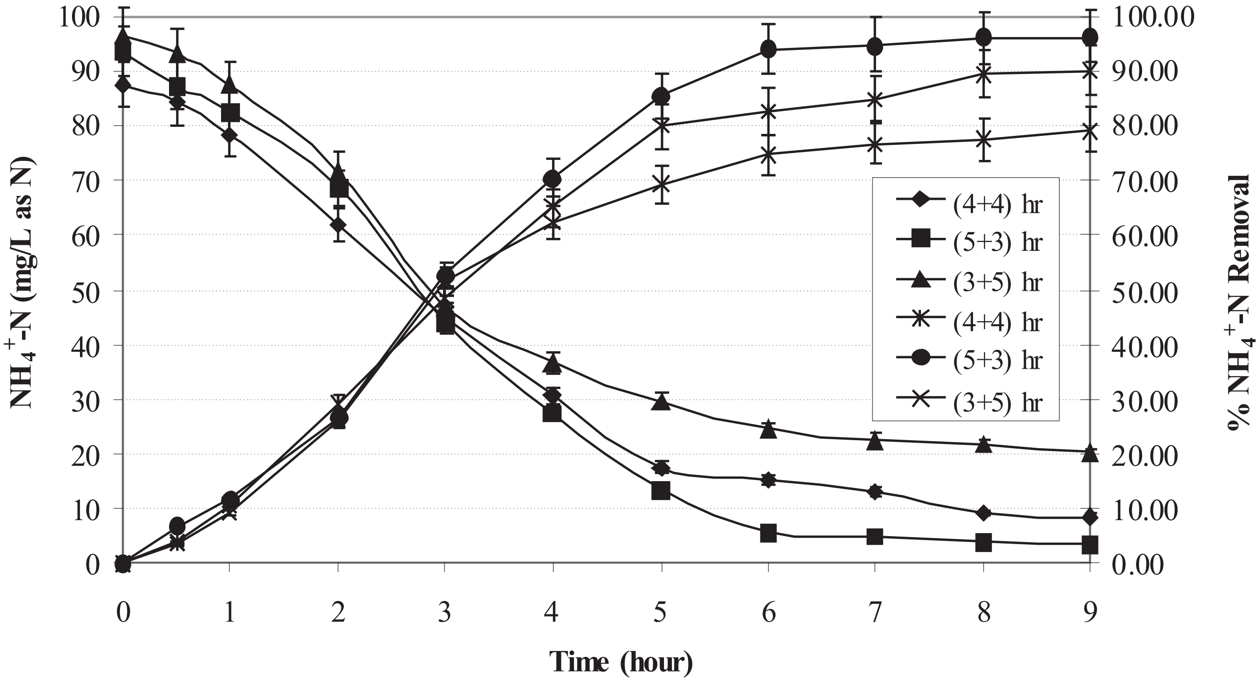

Figure 6: Ammonia oxidation profiles in SBR under different react period combination [initial SCOD = 1000 ± 50 mg/L and initial NH_4^+-N = 90 ± 10 mg/L as N].

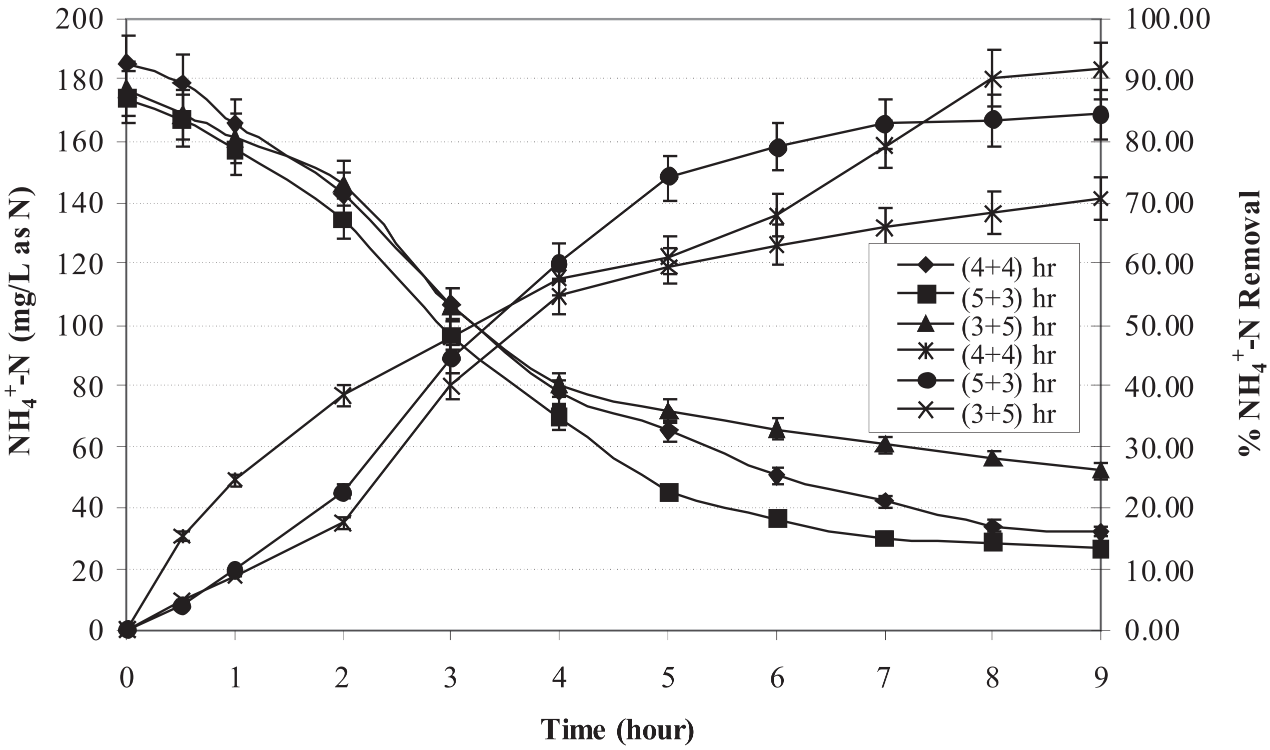

Figure 7: Ammonia oxidation profiles in SBR under different react period combination [initial SCOD = 2000 ± 50 mg/L and initial NH_4^+-N = 180 ± 10 mg/L as N].

Nitrite and Nitrate-Nitrogen Concentration Profile of Slaughterhouse Wastewater in SBR

The nitrite and nitrate-nitrogen (NO_2^--N and NO_3^--N) profile in the reactor is shown in Figure 8.The maximum nitrite level was observed in between 2.5 and 3.0 hour of react period. The peak nitrate (NO_3^-) level was formed close to 4.0 hours of aeration period for (4+4) and (3+5) hours combinations of react period. A time lag of one hour for maximum nitrate formation was noticed after the attainment of the maximum NO_2^--N level in the reactor. For (5+3) hour react period combination, the formation of NO_3^- showed a time dependent factor as the peak was found at the end of 5.0 hours. In Figure 8, after 4.0 hours of aeration period, the NO_3^- level was shown 35.21 mg/L as N corresponding to initial NH_4^+-N level of 87.52 mg/L as N and NO_3^- concentration of 12.35 mg/L as N, respectively. On the other hand, after 5.0 hours of aerated react period, NO_3^--N concentration in the reactor was found to be 60.24 mg/L as N for an initial NH_4^+-N and NO_3^--N concentration of 93.54 and 16.52 mg/L as N, respectively. The maximum NO_3^--N concentration for (3+5) hour react period combination was found to be 25.31 mg/L as N for the initial NH_4^+-N concentration of 96.58 mg/L as N and NO_3^--N level of 12.35 mg/L as N. The experimental results clearly indicate the necessity of longer aeration period for achieving maximum utilization of ammonia by the nitrifiers.

In Figure 9, after four hours of anoxic react period, nitrate (NO_3^-) was reduced to 22.29 mg/L as N from its peak concentration of 96.22 mg/L as N, which indicate 76.83% removal of nitrate for initial NH_4^+-N concentration 185.24 mg/L as N. During denitrification phase, the residual soluble COD concentration was found to be more than the stoichiometric organic carbon requirement for effective denitrification meeting all metabolic requirements. When the anoxic react period was changed to three hours, it has been observed that, though nitrate concentration after five hours of aerobic period was found to be maximum (92.11 mg/L as N), per cent removal of nitrate descended from 76.83 to 66.16% for initial NH_4^+-N concentration 173.88 mg/L as N, due to insufficient of anoxic period.

MLVSS, pH, Alkalinity and DO Profiles in the SBR during Experiment

In the present system a complex biomass culture co-existed in the reactor, where autotrophic organisms under aerobic conditions act during ammonia oxidation stage while the heterotrophic bacteria perform carbon oxidation. This nitrification step is followed by a denitrification that is carried by heterotrophic bacteria under anoxic conditions responsible for converting nitrate to molecular nitrogen. It has been observed that a steady increase in MLVSS level occurred progressively during experiment. The rate of increase in anoxic

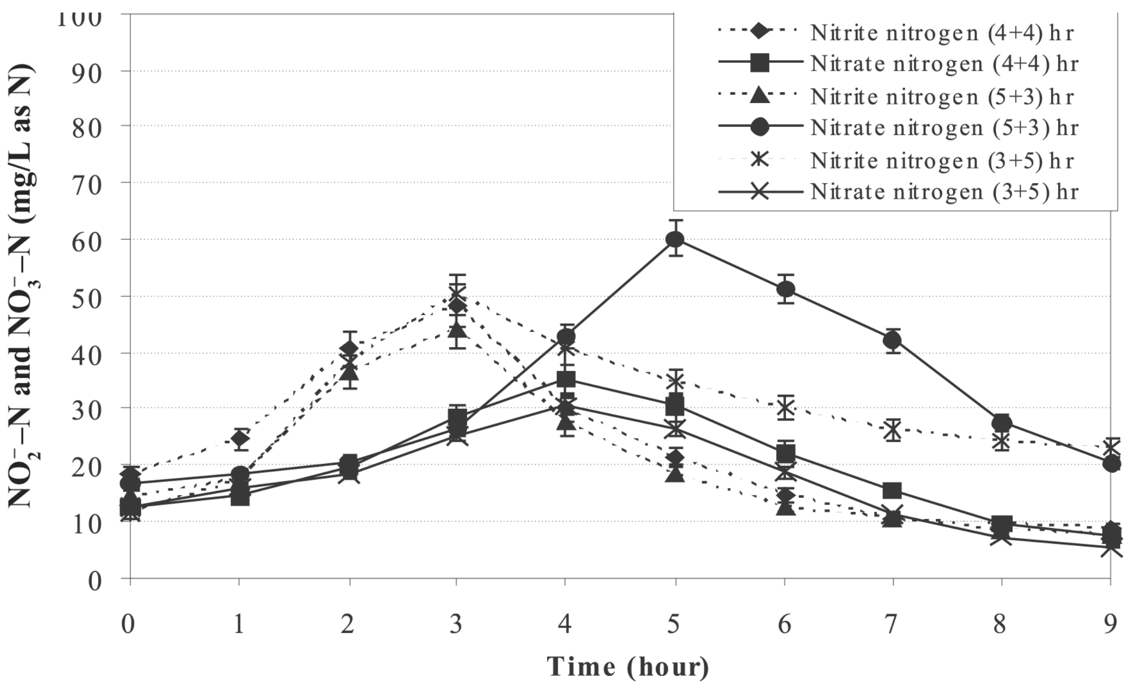

Figure 8: Nitrite and nitrate concentration profiles in SBR under different react period combination [initial SCOD = 1000 ± 50 mg/L and initial NH_4^+-N = 90 ± 10 mg/L as N].

period was found to be marginal than that observed during aerobic react period (Figures 10, 11 and 12). The reason behind this phenomenon can be explained on the basis of this fact that during anoxic react period facultative microbes were dominant and the growth of such microbes was not usually high as compared to the growth of aerobic heterotrophs. However, the overall increase in MLVSS in both the phases demonstrated

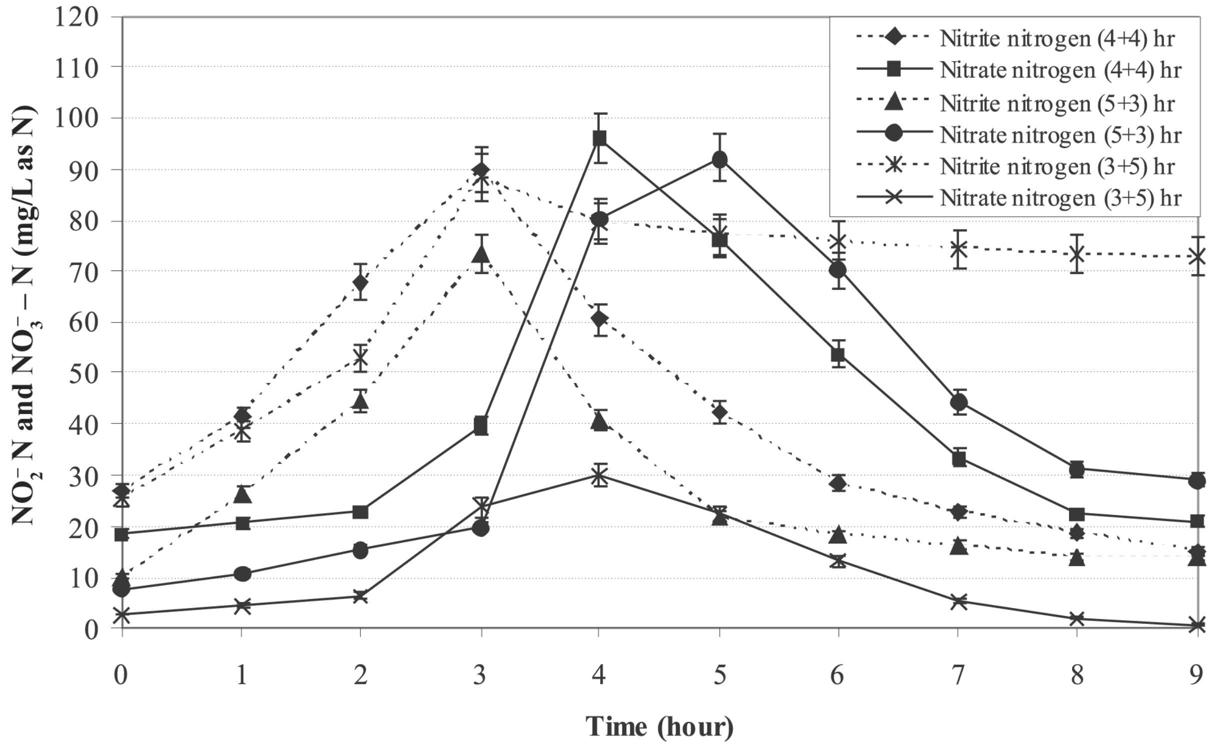

Figure 9: Nitrite and nitrate concentration profiles in SBR under different react period combination [initial SCOD = 2000 ± 50 mg/L and initial NH_4^+-N = 180 ± 10 mg/L as N].

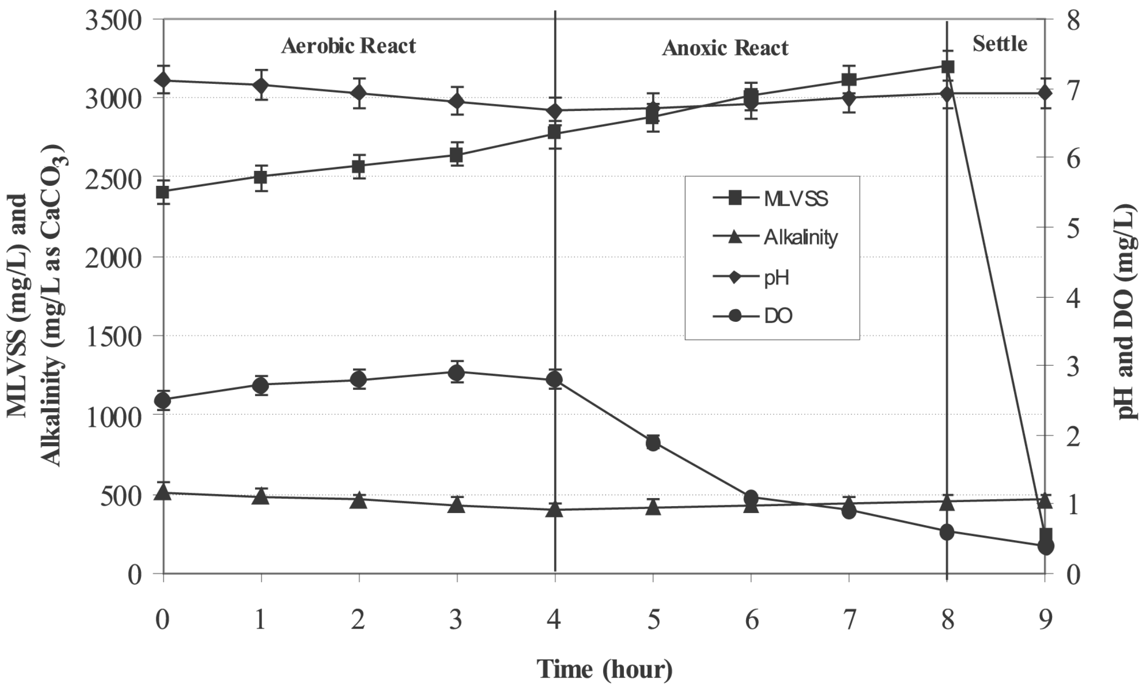

Figure 10: MLVSS, pH, alkalinity and DO profiles for slaughterhouse wastewater treatment in SBR under (4+4) hour react period combination.

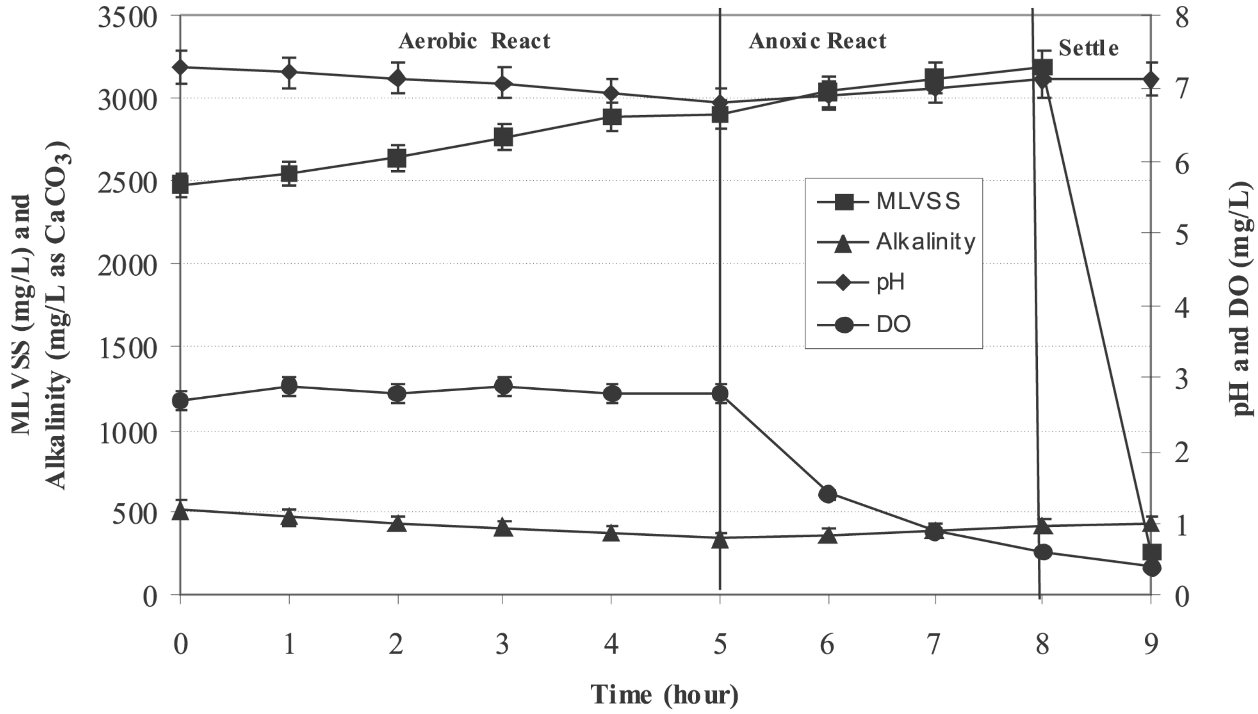

Figure 11: MLVSS, pH, alkalinity and DO profiles for slaughterhouse wastewater treatment in SBR under (5+3) hour react period combination.

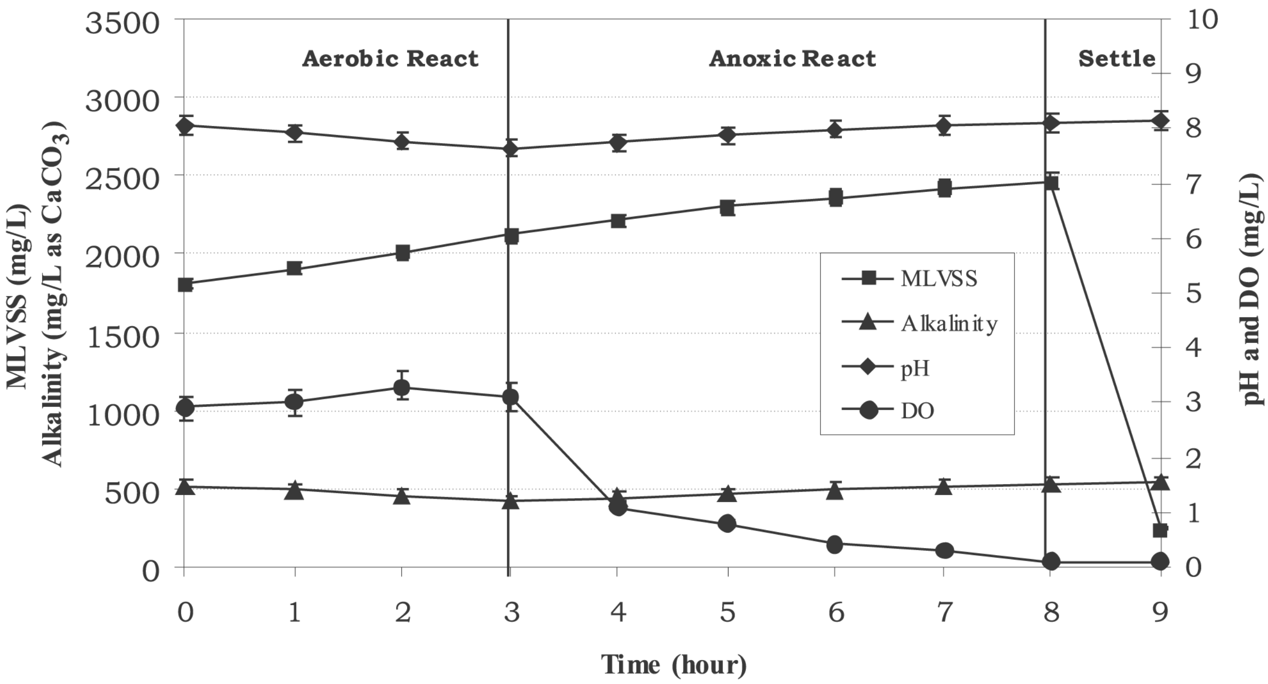

Figure 12: MLVSS, pH, alkalinity and DO profiles for slaughterhouse wastewater treatment in SBR under (3+5) hour react period combination.

the microbial activity for organic carbon oxidation, nitrification and denitrification.

The pH and alkalinity values of a biological system are vital parameters for microbial reactions, and hence, the variation in pH and alkalinity often provides a good indication of the ongoing biological reactions, e.g., increases in pH for ammonification and denitrification and decreases in pH owing to organic carbon oxidation and nitrification. Alkalinity is not only important for nitrification and denitrification; it can also be used to indicate the system stability. Alkalinity was found to have a close correlation with SBR operating conditions, since different extents of nitrification (alkalinity consumption) and denitrification (alkalinity production) contribute to the variation of alkalinity in the system. During the aerobic phase, the minima of the pH curve characterized the end of nitrification (Figures 10, 11 and 12). At the beginning of anoxic react phase, when ammonia-nitrogen concentration had reduced considerably, pH began to increase. This has occurred between 4.0 and 5.0 h after the commencement of aerobic stage in all sets.

In Figures 10, 11 and 12, the DO profiles exhibited a sharp bend after which DO concentration decreased markedly at anoxic phase and reached its minima. In the present study, the DO level remained almost steady during the entire aerobic react period with a marginal increase in DO level but a marked descending trend was observed during anoxic period in all the reaction sets irrespective of initial SCOD and ammonia concentrations. Under strict anaerobic condition the DO should be equal to zero but anoxic environment starts from DO level less than 1.5 mg/L. At the start of anoxic react period, in most of the cases, DO was found to be less than 1.5 mg/L and at the end of anoxic react period the value becomes less than 1.0 mg/L.

Conclusions

The following conclusions are drawn based on the present work on simultaneous removal of organic carbon and nitrogen from slaughterhouse wastewater in a laboratory scale SBR:

1. The SCOD removal during aerobic react period was achieved due to utilization of substrate as required of aerobic microorganisms in the mixed culture. In the anoxic phase, the residual SCOD was utilized by the facultative microbes.
2. It is observed that the SBR can perform efficiently in achieving nitrification and denitrification sequentially along with oxidation of organic carbon. The combination of 4-hour aerobic react period and 4-hour anoxic react period has been found to be optimum from the view point of both nitrification and denitrification.
3. Length of aeration time in 5-hr (aerobic) and 3-hr (anoxic) mode react phase of the operation cycle, did not have any significant impact on SCOD reduction.
4. Longer aeration period (five hours) has been found to be effective in achieving higher degree of nitrification. However, it affected the percent removal of nitrate due to the prevalence of shorter anoxic period essential for denitrification.
5. The pH level in the SBR descended initially during aerobic period due to nitrification and carbon oxidation followed by an increasing trend indicating the existence of denitrifiers. This phenomenon has also been established by the variation of alkalinity level during aerobic and anoxic react period.
6. A steady increase in MLVSS level was occurred progressively during experiment. The overall increase in MLVSS in both the phases demonstrated the microbial activity for organic carbon oxidation, nitrification and denitrification.

Acknowledgement

This study was supported by the research funds of Jadavpur University, Jadavpur, Kolkata – 32.

References

Al-Mutairi, N.Z., Hamoda, M.F. and I.A. Al-Ghusain (2007). Slaughterhouse wastewater treatment using date seeds as adsorbent. *Journal of Environment Science and Health*, **55:** 678-710.

Al-Mutairi, N.Z., Al-Sharifi, F.A. and S.B. Al-Shammari (2008). Evaluation study of a slaughterhouse wastewater treatment plant including contact-assisted activated sludge and DAF. *Desalination*, **225:** 167-175.

American Public Health Association (APHA) (1991). Standard Methods for the Examination of Water and Wastewater (22^{nd} edn). APHA, AWWA, WPCF, Washington, D.C.

Boopathy, R., Bonvillain, C., Fontenot, Q. and M. Kilgen (2007). Biological treatment of low-salinity shrimp aquaculture wastewater using sequencing batch reactor. *International Biodeterioration and Biodegradation*, **59:** 16-19.

Cassidy, D.P. and E. Belia (2005). Nitrogen and phosphorus removal from an abattoir wastewater in a SBR with aerobic granular sludge. *Water Research*, **39:** 4817-4823.

Del Pozo, R. and V. Diez (2005). Integrated anaerobic aerobic fixed-film reactor for slaughterhouse wastewater treatment. *Water Research*, **39:** 1114-1122.

Kargi, F. and A. Uygur (2002). Nutrient Removal Performance of a Sequencing Batch Reactor as a Function of the Sludge Age. *Enzyme Microbial. Technology*, **31:** 842-847.

Kern, C. and R. Boopathy (2012). Use of sequencing batch reactor in the treatment of shrimp aquaculture wastewater. *Journal of Water Sustainability*, **2(4):** 221-232.

Kim, H.S., Choung, Y.K., Ahn, S.J. and H.S. Oh (2008). Enhancing nitrogen removal of piggery wastewater by membrane bioreactor combined with nitrification reactor. *Desalination*, **223:** 194-204.

Lemaire, R., Yuan, Z., Nicolas, B., Marcelino, M., Yilmaz, G. and J. Keller (2009). A sequencing batch reactor system for high-level biological nitrogen and phosphorus removal from abattoir wastewater. *Biodegradation*, **20(3):** 339-350.

Li, J.P., Healy, M.G., Zhan, X.M. and M. Rodgers (2006). Nutrient Removal from Slaughterhouse Wastewater in an Intermittently Aerated Sequencing Batch Reactor. *Bioresource Technology*, **52:** 163-167.

Madigan, M.T., Martinko, J.M. and J. Parker (2000). Brock Biology of Microorganisms. 8th ed. Prentice-Hall, Englewood Cliffs, N.J.

Mahvi, A.H., Mesdaghinia, A.R. and F. Karakani (2004). Nitrogen Removal from Wastewater in a Continuous Flow Sequencing Batch Reactor. *Pakistan Journal of Biological Sciences*, **7(11):** 1880-1883.

Mittal, G.S. (2006). Treatment of wastewater from abattoirs before land application—A review. *Bioresour Technology*, **97:** 1119-1135.

Painter, H.A. (1977). Microbial Transformations of Inorganic Nitrogen. *Progress in Water Technology*, **8:** 3-29.

Palatsi, J., Vinas, M., Guivernau, M., Fernandez, B. and F. Flotats (2011). Anaerobic digestion of slaughterhouse waste: Main process limitations and microbial community interactions. *Bioresource Technology*, **102:** 2219-2227.

Rajagopol, P., Rousseau, Bernet, N. and F. Béline (2011). Combined anaerobic and activated sludge anoxic/oxic treatment for piggery wastewater. *Bioresourse Technology*, **102:** 2185-2192.

Roy, D., Hassan, K. and R. Boopathy (2010). Effect of carbon to nitrogen (C:N) ratio on nitrogen removal from shrimp production waste water using sequencing batch reactor. *Journal* of *Industrial Microbiology and Biotechnology*, **37:** 1105-1110.

Wang, Xiao, Zhi, F.Q., Zhi, Xu, Yan, G.H. and Z.S. Tong (2009). Study on Treatment of Slaughterhouse Wastewater with Co-coagulation Flotation and SBR Process. Xiaoweiwang127@163.com.

Wang, F., Liu, Yi, Wang, J., Zhang, Y. and H. Yang (2012). Influence of growth manner on nitrifying bacterial communities and nitrification kinetics in three lab-scale bioreactors. *Journal of Industrial Microbiology and Biotechnology*, **39:** 595-604.