

Performance Study and Kinetic Parameter Evolution of Mixed Microbial Consortia for Removal of Organic Carbon and Nitrogen from Wastewater Through Batch Test

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Abstract - In this study, biological treatment of waste water was carried out by mixed culture in a laboratory-scale batch reactor for combined removal of soluble organics and nitrogen using wastewater feed solution. The study include the preparation of acclimatized seeds for SCOD (soluble chemical oxygen demand) removal, ammonia nitrogen ($\text{NH}_4^+\text{-N}$) oxidation, nitrate nitrogen ($\text{NO}_3^-\text{-N}$) reduction and finally an investigation was carried out for removal of SCOD, ammonia nitrogen ($\text{NH}_4^+\text{-N}$) and nitrate nitrogen ($\text{NO}_3^-\text{-N}$) in a batch fed reactor using the mixed culture and subsequently estimated the bio-kinetic coefficient such as maximum rate of substrate utilization (k), Half saturation concentration (K_s), yield coefficient (Y), decay coefficient (k_d) using the batch experimental data. Various input combinations were tried on the basis of different proportion of initial substrate concentration (SCOD, $\text{NH}_4^+\text{-N}$ and $\text{NO}_3^-\text{-N}$) along with different mixed liquor suspended solid (MLVSS) concentration. It was observed that removal of 94.53 to 97.39% and 82.93 to 86.05% of SCOD was occurred after 48 hr for all experimental sets having initial SCOD concentration of 500 ± 50 mg/L and 1000 ± 50 mg/L respectively. Similar removal trend of 85.30 to 93.13% and 81.56 to 87.08% of $\text{NH}_4^+\text{-N}$ with respect to time was also observed after 48 hr for all sets of experiments corresponding to initial $\text{NH}_4^+\text{-N}$ concentration of 90 ± 10 mg/L and 140 ± 10 mg/L respectively. Almost 80.64 to 90.59% and 76.91 to 85.47% of $\text{NO}_3^-\text{-N}$ removal was found after 48 hr for all experimental sets having initial $\text{NO}_3^-\text{-N}$ concentration of 40 ± 10 mg/L and 90 ± 10 mg/L respectively.

Keywords:- *Biological Wastewater Treatment; Carbon Oxidation; Nitrification Denitrification; Kinetic Coefficient.*

1. INTRODUCTION

The characteristic of wastewater is an important factor for the design and operation of wastewater treatment facilities. The properties and composition of wastewater

depends primarily on the source of generation. Traditionally wastewater treatment has focused on the removal of gross organic and inorganic constituents and pathogens in wastewater that primarily included carbonaceous BOD and suspended solids removal and disinfection processes. Nitrogen was also considered as a significant problem. Organic carbon and nitrogen have become contaminants of concern in wastewater. Nitrogen is essential nutrient for growth. But due to the high level of nitrogen, present in the treated effluent to be discharged into the surface water is not desirable since they are potentially hazardous both for aquatic and human life. They lead to some undesirable problems. The various forms of nitrogen as nitrate, nitrite, organic nitrogen, and ammonia cause several problems. Being stable and highly soluble, nitrate serves as algal nutrients. Nitrogen is major component responsible for eutrophication. Algal blooms create unaesthetic conditions. Nitrates may also lead to the possible formation of nitrosoamines, which are known as carcinogens. Hence treatment is necessary for the removal of organic carbon and nitrogen which are very harmful to the environment.

There are several treatment options for the treatment of wastewater containing organic carbon and nitrogens, such as physical, chemical, physico-chemical and biological treatment. Out of these, biological treatment is the best choice owing to its low cost, reliable and effective nature [1]. The efficiency of nutrient removal is also very effective in biological treatment. According to Kargi and Dincer [2] biological treatment is the best alternative to treat high strength wastewater containing carbonaceous organic matter and nutrients. The main purpose of biological wastewater treatment is to reduce the biological oxygen demand in the wastewater through oxidization of organic matter to carbon dioxide and water. A large number of bacteria oxidize the

organic matter and can remove nitrogen from the wastewater. Biological treatment of wastewater has been employed successfully for many types of industries and commonly used as a secondary treatment.

The performance of biological process or treatment of wastewater depends on the dynamic of substrate utilization and microbial growth kinetics. Kinetic parameters determination is very important features for the design of suitable wastewater treatment. The kinetic constants such as half saturation concentration (K_s), maximum rate of substrate utilization (k), yield coefficient (Y) and endogenous decay coefficient (K_d) are to be determined. These co-efficient values are used to predict the rate of substrate utilization and biomass growth can vary as a function of the wastewater source, microbial population and temperature. The necessity of determining kinetics in a batch fed reactor is to improve the design and operation of wastewater treatment plants and to compare the kinetics value of real life wastewater sample with that of synthetic wastewater.

Biological treatment of wastewater containing organic carbon and nitrogen (COD and TKN) are also carried out in laboratory and pilot scale experiment by several researchers successfully [3-11]. Nutrients in piggery wastewater with high organic matter, nitrogen and phosphorous content were biological removed by Obaja et al. [12] in a sequencing batch reactor (SBR) with anaerobic, aerobic and anoxic stages. The SBR was operated with wastewater containing 1500 mg/L ammonium and 144 mg/L phosphate, a removal efficiency of 99.7 % for nitrogen and 97.3 % for phosphate was obtained. A full scale SBR system was evaluated by Lo and Liao [13] to remove 82 % of BOD and more than 75% of nitrogen after a cycle period of 4.6 hour from swine wastewater. Mahvi et al. [14] 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 objective of the present investigation was to carry out a performance evaluation of a laboratory-scale batch reactor for combined removal of soluble organics and nitrogen using wastewater feed solution and evaluate the bio-kinetic coefficients for carbon oxidation, nitrification and denitrification under different initial SCOD, initial ammoniacal nitrogen ($\text{NH}_4^+\text{-N}$) and initial nitrate nitrogen ($\text{NO}_3^-\text{-N}$) concentration.

2. MATERIAL AND METHODS

2.1 SEED ACCLIMATIZATION

The microbial seeds were acclimatized under laboratory condition for simultaneous process of carbon oxidation, nitrification and denitrification. It was conducted in a measuring cylinder of 2.0 L capacity. A separate seed acclimatization programme was also performed to develop carbon utilizing heterotrophic microbes and autotrophic nitrifiers. The active microbial seed was cultured under ambient condition in the laboratory by inoculating 750mL sludge as collected from nearby sewage treatment plant, to a growth propagating media composed of 500 mL dextrose solution having concentrations of 1000 mg/L and 50 mL of nutrient solution in the above cylinder. Aeration was done

by means of diffused air system using two numbers of aquarium pumps. Only dextrose ($\text{C}_6\text{H}_{12}\text{O}_6$) was added intermittently as a carbon source. The composition of nutrients and trace element has been given in Table-I. The acclimatization process was continued for a period of three months. The biomass growth was monitored by the magnitude of sludge volume index (SVI) and MLVSS concentration. pH in the reactor was maintained in the range of 6.8-7.0 by adding required amount of sodium carbonate (Na_2CO_3) and phosphate buffer. Similarly for nitrification, the seed was separately prepared using 250 mL ammonium chloride (NH_4Cl) solution as substrate having concentration of 200 mg/L along with other macro and micronutrients and trace elements. The acclimatization was carried out over a period of three months; pH was adjusted in the range of 7.2-8.0 by adding sodium carbonate solution (2%).

The concentrated biomass including nitrifiers then mixed in a separate glass cylinder, to examine the combined carbon oxidation and nitrification behavior of mixed culture process. SCOD in the form of dextrose of strength 1000 mg/L and ammonium chloride of strength 200mg/L were used as sources of carbon and ammonia nitrogen ($\text{NH}_4^+\text{-N}$) respectively. Nutrients and trace materials were added as per recommended dosage and as exhibited in Table-I. Air was supplied by means of diffused air system. 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. Combined carbon oxidation and nitrification study was carried out by varying dextrose dose from 500 ± 50 to 1000 ± 50 mg/L with ammonia nitrogen solution varying from 40 ± 10 to 180 ± 10 mg/L as N. The seed acclimatization phase was stopped when a steady state performance were observed in terms of equilibrium SCOD and $\text{NH}_4^+\text{-N}$ reduction with respect to a steady level of MLVSS concentration and SVI in the reactor.

Microbial seed for denitrification study developed separately in 2.0 L capacity aspirator bottle. About 500 gm of digested sludge obtained from an anaerobic chamber in a slaughterhouse waste treatment plant was added to 1.0 L of distilled water. The solution was filtered and the resulting solution was acclimatized for denitrification purpose using dextrose as carbon source (1000 mg/L) and potassium nitrate (20 mg/L as N) as the source of nitrate nitrogen ($\text{NO}_3^-\text{-N}$). Magnetic stirrer was provided for proper mixing of the solution. The consortium was acclimatized gradually to higher concentrations of nitrate up to 90mg/L as N towards the end of the acclimatization period, over a span of 90 days. The pH in the reactor was maintained in the range of 6.8-8 depending upon the anoxic/anaerobic condition. The seed acclimatization phase was continued until a steady state condition was observed that is equilibrium SCOD and nitrate removal vis a vis MLVSS concentration.

TABLE-I: COMPOSITION OF NUTRIENTS AND TRACE ELEMENTS

Nutrients	Mass to be added in 1000mL, in mg
K ₂ HPO ₄	60.0
KH ₂ PO ₄	40.0
Trace element	Mass to be added in 1000mL, in mg
MgSO ₄ .7H ₂ O	500.0
FeCl ₃ .6H ₂ O	710.0
ZnSO ₄ .7H ₂ O	0.1
CuSO ₄ .5H ₂ O	0.1
MnCl ₂ .2H ₂ O	8.0
(NH ₄) ₆ Mo ₇ O ₂₄	0.11
CaCl ₂ .2H ₂ O	100.0
CoCl ₂ .6H ₂ O	200.0
Al ₂ (SO ₄) ₃ .16H ₂ O	55.0
H ₃ BO ₃	150.0
EDTA	100.0

2.2 EXPERIMENTAL SETUP AND PROCEDURE

Batch kinetic studies for treatment of synthetically prepared wastewater for carbon oxidation, nitrification and simultaneous carbon oxidation and ammonia nitrification have been performed in 2L volume measuring cylinder separately as shown in Fig.-1. About 1.5 L volume of sample was taken in each cylinder with necessary feed solution. Each cylinder was facilitated with aeration system by inserting stone sparger fitted with laboratory scale compressor. The graduation marked in the cylinder helped for determining sludge volume for each cylinder in more convenient manner.

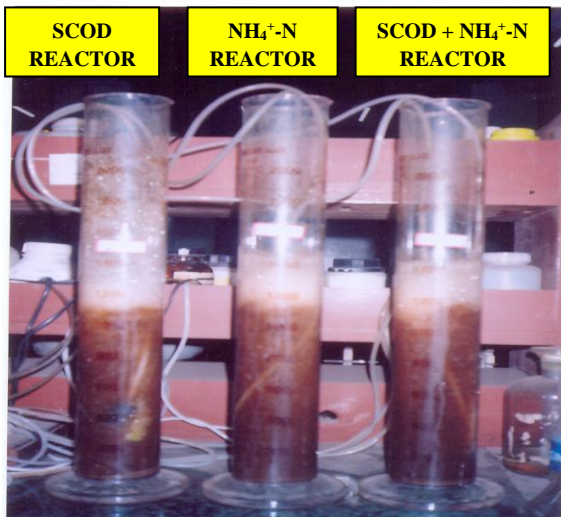


Fig.-1: Experimental Setup of Batch Reactor for Carbon oxidation and Nitrification Study

2.3 CARBON OXIDATION STUDY IN BATCH REACTOR

A mixed liquor of 100ml, containing mixed carbonaceous sludge as acclimatized previously, was taken into 2.0 L volume of batch reactor. The seed was acclimatized in new environment to a feed having SCOD around 500-1000 mg/L by adding dextrose (C₆H₁₂O₆) as carbon source for a period of two weeks. Aeration was done continuously by means of diffused air systems, by aqua pumps. During this acclimatization process a healthy

biomass growth has been observed. The range of pH value of this synthetically prepared wastewater was maintained as 7.0 to 7.5 by adding sodium carbonate solution (2%) at required amount. During the time course of entire kinetic study of the carbon oxidation, a 50ml sample were separately collected from the reactor at different time interval Viz. 2, 4, 6, 8, 10, 24, 48 hrs for both combination-A and combination-B. The operation conditions for both combination-A and combination-B in carbon oxidation study have been exhibited in Table-II and Table-III, respectively.

TABLE-II: COMBINATION-A (INITIAL SCOD CONCENTRATION 500±50 mg/L)

Set Number	Initial COD Concentration (mg/L)	Initial MLVSS (mg/L)	Initial pH	Initial DO (mg/L)
Set-1	475.23	173.38	8.0	2.6
Set-2	464.55	188.22	7.8	2.8
Set-3	542.33	221.52	7.6	3.1
Set-4	520.22	209.81	8.0	2.8

TABLE-III: COMBINATION-B (INITIAL SCOD CONCENTRATION 1000±50 mg/L)

Set Number	Initial COD Concentration (mg/L)	Initial MLVSS (mg/L)	Initial pH	Initial DO (mg/L)
Set-5	972.56	425.41	8.1	3.2
Set-6	1011.23	422.53	7.9	3.4
Set-7	1036.42	435.24	7.7	2.9
Set-8	1046.52	437.52	7.8	3.5

2.4 NITRIFICATION STUDY IN BATCH REACTOR

Similarly for nitrification, a mixed liquor of 100ml, containing mixed nitrogenous sludge as previously acclimatized, was taken into a 2.0 L volume in a batch reactor. The seed was acclimatized in the new environment against a feed having NH₄⁺-N concentration around 90-140 mg/L by adding ammonium chloride (NH₄Cl) as nitrogen source for a period of two weeks. Aeration was continuously done by means of diffused air systems. During this acclimatization process an active biomass growth was developed. The range of pH value of this synthetically prepared wastewater was maintained as 7.0 to 7.5 by adding sodium carbonate solution (2%) at required amount. During the course of entire kinetic study of nitrification study a 50ml sample were collected from the reactor at different time interval Viz. 2, 4, 6, 8, 10, 24, 48 hrs for both combination-C and combination-D. The operation conditions for both combination-C and combination-D in nitrification study are shown in Table-IV and Table-V, respectively.

TABLE-IV: COMBINATION-C (INITIAL NH₄⁺-N CONCENTRATION 90±10 mg/L)

Set Number	Initial NH ₄ ⁺ -N Concentration (mg/L)	Initial MLVSS (mg/L)	Initial pH	Initial DO (mg/L)
Set-1	94.50	112.52	7.8	3.0
Set-2	82.16	108.52	7.5	2.5
Set-3	98.33	120.58	7.9	2.8
Set-4	86.22	116.58	7.6	2.6

TABLE-V: COMBINATION-D (INITIAL NH₄⁺-N CONCENTRATION 140±10 mg/L)

Set Number	Initial NO ₃ ⁻ -N Concentration (mg/L)	Initial MLVSS (mg/L)	Initial pH	Initial DO (mg/L)
Set-5	96.82	118.75	8.1	1.2
Set-6	92.26	126.55	7.6	1.1
Set-7	86.63	110.26	7.9	1.1
Set-8	88.35	128.54	8.0	1.3

2.5 DENITRIFICATION STUDY IN BATCH REACTOR

Denitrification study was performed separately in 2.0 L aspirator bottle as shown in Fig.-2. A mixed liquor of 100ml, containing previously acclimatized denitrifying sludge, was taken into a 2.0 L volume of aspirator bottle. The seed was acclimatized in new environment to a feed having NO₃⁻-N concentration around 40-90 mg/L by adding potassium nitrate (KNO₃) as nitrogen source for a period of two weeks. Magnetic stirrer was provided for proper mixing of the reactor solution. The range of pH value was maintained as 7.0 to 8.0 by adding sodium carbonate solution (2%) at required amount. During the time course kinetic study of denitrification, 50ml sample were collected from the aspirator bottle at different time interval Viz. 2, 4, 6, 8, 10, 24, 48 hrs for both combination-E and combination -F. The operation conditions for both combination-E and combination -F in denitrification study are shown in Table-VI and Table-VII, respectively.



Fig.-2: Experimental Setup of Batch Reactor for Denitrification Study

TABLE-VI: COMBINATION-E (INITIAL NO₃⁻-N CONCENTRATION 40±10 mg/L)

Set Number	Initial NO ₃ ⁻ -N Concentration (mg/L)	Initial MLVSS (mg/L)	Initial pH	Initial DO (mg/L)
Set-1	43.51	98.56	8.2	1.1
Set-2	42.26	105.84	7.8	1.2
Set-3	48.32	112.35	7.7	1.3
Set-4	36.25	95.35	8.0	1.2

TABLE-VII: COMBINATION-F (INITIAL NO₃⁻-N CONCENTRATION 90 ± 10 mg/L)

Set Number	Initial NH ₄ ⁺ -N Concentration (mg/L)	Initial MLVSS (mg/L)	Initial pH	Initial DO (mg/L)
Set-5	146.77	158.57	8.1	3.2
Set-6	142.56	145.57	7.7	3.4
Set-7	136.63	138.57	7.8	3.1
Set-8	148.32	165.22	8.0	3.5

2.6 CARBON OXIDATION AND NITRIFICATION KINETICS

Bio-kinetic parameters play an important role in designing and optimizing an activated sludge process. The bio-kinetic constants describe the metabolic performance of the microorganisms when subjected to the substrate and other components of the specific wastewater. These bio-kinetic coefficients yield a set of realistic design parameters, which can be used in rationalizing the design of the activated sludge process for a specific substrate.

a) Substrate removal kinetics

The substrate removal constants namely half saturation concentration (K_s) and the maximum rate of substrate utilization (k) were determined from the Lawrence and McCarty's modified Monod equation [15] given below:

$$1/U = (K_s/k)(1/S) + 1/k \text{ -----(1)}$$

S = Substrate (SCOD or NH₄⁺-N or NO₃⁻-N) concentration at any time in reactor (mg/L)

U = Specific substrate utilization rate = (S₀-S)/θX (mg of SCOD or mg of NH₄⁺-N or mg of NO₃⁻-N /day/mg of MLVSS)

θ = Contact time (day)

X = MLVSS at any time in the reactor (mg/L)

S₀ = Substrate (SCOD and NH₄⁺-N) concentration of the influent (mg/L)

The plots made between 1/U and 1/S develops into a straight line with K_s/k as its slope and 1/k as its intercept.

b) Sludge growth kinetics

The sludge growth kinetic constants namely the yield coefficient (Y) and the endogenous decay coefficient (K_d), were determined from the Lawrence and McCarty's modified Monod equation [15] given below:

$$1/\theta = YU - K_d \text{ ----- (2)}$$

Where U = Specific substrate utilization rate (mg of SCOD or mg of NH₄⁺-N /day/mg of MLVSS)

θ = Contact time (day)

k_d = Endogenous decay coefficient (per day)

$Y =$ Yield coefficient (mg of MLVSS produced/mg of SCOD or $\text{NH}_4^+\text{-N}$)
 A graph drawn between $1/\theta$ and U gives a straight line, with Y as its slope and k_d as its intercept.

3. RESULTS AND DISCUSSIONS

3.1 CARBON OXIDATION STUDY IN BATCH REACTOR

The results of carbon oxidation study for combination-A having initial SCOD concentration of 500 ± 50 mg/L and combination-B having initial SCOD concentration of 1000 ± 50 mg/L are shown in Fig.-3 and Fig.-4 respectively. From Fig.-3 it is observed that the initial concentration of SCOD decreases with the progress of reaction time. The Fig.-6.1 also shows that within 10 hr of contact time maximum SCOD removal was achieved beyond which removal percentage is found to be marginal and the curve becomes asymptotic in nature which indicated maximum stabilization of biodegradable portion of the waste within the reaction period.

Similar removal trend with respect to time is also observed in Fig.-4 with respect to time irrespective of any initial SCOD value in the range of 1000 ± 50 mg/L. Almost 94.53 to 97.39% and 82.93 to 86.05% of SCOD removal occurred after 48 hr for all of the experimental sets having initial SCOD concentration of 500 ± 50 mg/L (combination-A) and 1000 ± 50 mg/L (combination-B) respectively. The results reveal that the kinetic reactions are not as such influenced by initial SCOD loading up to a value of 1000 ± 50 mg/L, as such. No substrate inhibition was observed in the present experimental set as evident in Fig.-3 and Fig.-4. SCOD level was decreased rapidly during the aerobic phase by the heterotrophic carbon oxidizing bacteria through the process of satisfying their metabolic energy requirement and cell synthesis substrate demand.

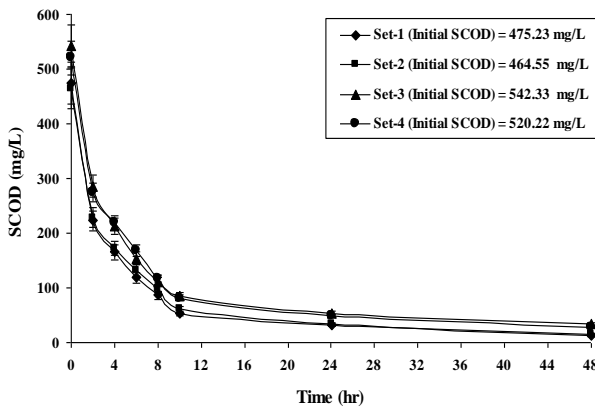


Fig.-3: SCOD Removal Profile during Carbon Oxidation Study in Batch Reactor [Initial SCOD Concentration of 500 ± 50 mg/L]

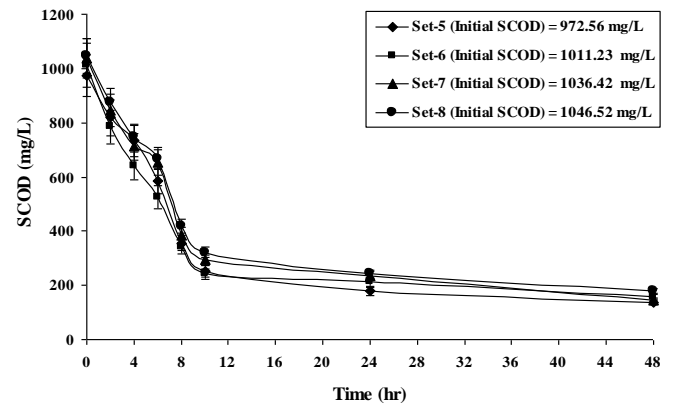


Fig.-4: SCOD Removal Profile during Carbon Oxidation Study in Batch Reactor [Initial SCOD Concentration of 1000 ± 50 mg/L]

3.2 NITRIFICATION STUDY IN BATCH REACTOR

The results of batch nitrification study for combination-C having initial $\text{NH}_4^+\text{-N}$ concentration of 90 ± 10 mg/L and combination-D having initial $\text{NH}_4^+\text{-N}$ concentration of 140 ± 10 mg/L have been plotted in Fig.-5 and Fig.-6 respectively. From both the figures, it is observed that as the time increases the $\text{NH}_4^+\text{-N}$ concentration found decreases and ultimately after 48 hr, the residual $\text{NH}_4^+\text{-N}$ concentration inside the reactor ceased to a minimal level. Fig.-5 and Fig.-6 also exhibit that within 10 hr, maximum $\text{NH}_4^+\text{-N}$ was oxidized. During these periods, the biological nitrification took place through oxidation of ammonia, by the activity of nitrifiers. Similar removal trends are observed in the present study with respect to time irrespective of any initial $\text{NH}_4^+\text{-N}$ concentration values over the range of 90 ± 10 to 140 ± 5 mg/L. Almost 85.30 to 93.13% and 81.56 to 87.08% of $\text{NH}_4^+\text{-N}$ removal occurred after 48 hr for all of the experimental sets corresponding to initial $\text{NH}_4^+\text{-N}$ concentration of 90 ± 10 mg/L (combination-C) and 140 ± 10 mg/L (combination-D) respectively. The higher rate of removal of ammonia nitrogen took place perhaps due to the assimilation of nitrogenous substrate by the nitrifiers.

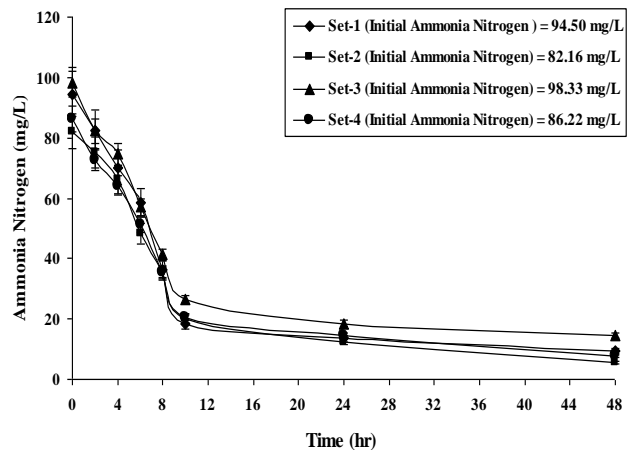


Fig.-5: Ammonia Nitrogen ($\text{NH}_4^+\text{-N}$) Removal Profile during Nitrification Study in Batch Reactor [Initial $\text{NH}_4^+\text{-N}$ Concentration of 90 ± 10 mg/L]

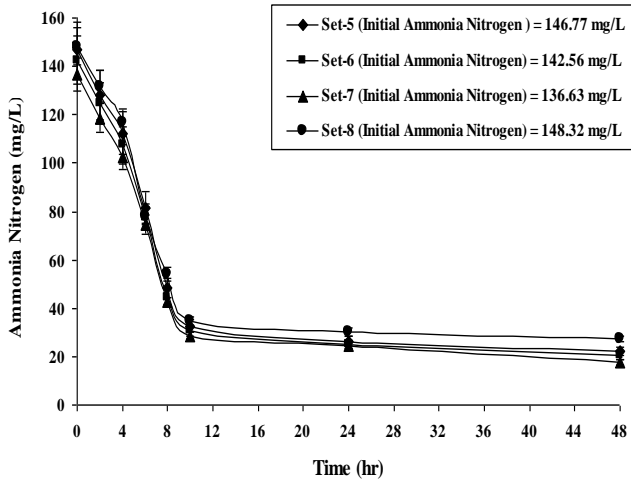


Fig.-6: Ammonia Nitrogen ($\text{NH}_4^+\text{-N}$) Removal Profile during Nitrification Study in Batch Reactor [Initial $\text{NH}_4^+\text{-N}$ Concentration of 140 ± 10 mg/L]

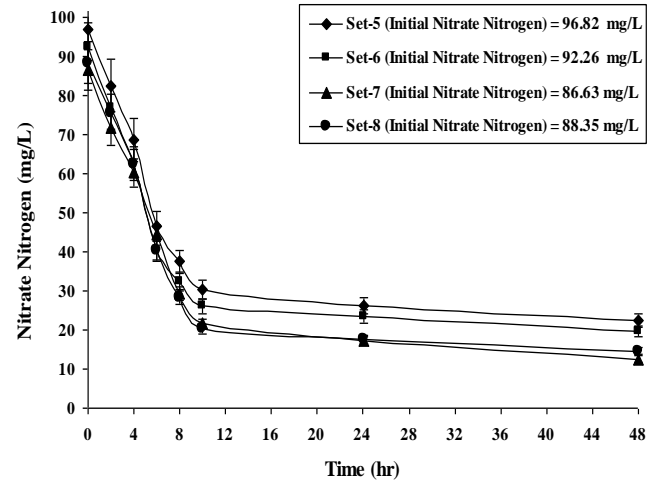


Fig.-8: Nitrate Nitrogen ($\text{NO}_3^-\text{-N}$) Removal Profile during Denitrification Study in Batch Reactor [Initial $\text{NO}_3^-\text{-N}$ Concentration of 90 ± 10 mg/L]

3.3 DENITRIFICATION STUDY IN BATCH REACTOR

The results of denitrification study for combination-E having initial $\text{NO}_3^-\text{-N}$ concentration of 40 ± 10 mg/L and combination-F having initial $\text{NO}_3^-\text{-N}$ concentration of 90 ± 10 mg/L are shown in Fig.-7 and Fig.-8 respectively. From Fig.-7 and Fig.-8, it is observed that the initial concentration of $\text{NO}_3^-\text{-N}$ decreases with the progress of time and maximum $\text{NO}_3^-\text{-N}$ was removed within a period of 10 hr of contact period. Similar removal trend is observed irrespective of initial $\text{NO}_3^-\text{-N}$ concentration values over the range of 40 ± 10 to 90 ± 10 mg/L. The removal patterns for $\text{NO}_3^-\text{-N}$ from the solution under anoxic condition is found to be high till a period of 10 hr, after which the removal was marginal. Initially, high organic carbon level in the system satisfies the organic carbon required by the denitrifiers. As the organic carbon level, measured as COD, decreased in the latter phase the activity of denitrifiers also descended, and consequently the rate of denitrification also found to be reduced at end of reaction. Almost 80.64 to 90.59% and 76.91 to 85.47% of $\text{NO}_3^-\text{-N}$ removal occurred after 48 hr for all of the experimental sets having initial $\text{NO}_3^-\text{-N}$ concentration of 40 ± 10 mg/L (combination-E) and 90 ± 10 mg/L (combination-F) respectively.

3.4 KINETICS FOR CARBON OXIDATION

The values for the reciprocal of specific substrate utilization rate ($1/U_C$) were plotted against the reciprocal of effluent SCOD ($1/S$) and substrate removal kinetics was evaluated using simple linear equation $1/U_C = (K_s/k)(1/S) + 1/k$. A best fit graph was drawn by applying a least square approach using experimental data as shown in Fig.-9 and Fig.-10. The slope and intercept of the straight line are K_s/k and $1/k$ respectively. The values of k were 5.05 per day and 4.87 per day for initial SCOD concentration of 500 ± 50 mg/L and 1000 ± 50 mg/L respectively. The maximum substrate utilization rate (k) decreased slightly under high initial SCOD concentration of 1000 ± 50 mg/L, showing a gradual build up of substrate inhibition for heterotrophs within the batch reactor. The half velocity constant (K_s) was increased appreciably from 243.05 to 303.23 mg/L by doubling of input substrate level from 500 ± 50 to 1000 ± 50 mg/L respectively. The value of K_s obtained for carbon-oxidation was found to be higher than the standard value of kinetics coefficient of municipal wastewater treatment [1] due to higher initial concentrations of SCOD in the batch reactor. Furthermore, the values of the reciprocal of the reaction time ($1/\theta$) were plotted against specific substrate utilization rate (U_C) as shown in Fig.-11 and Fig.-12. The yield coefficient (Y) was determined from the slope of the best-fit straight line and endogenous decay coefficient (K_d) was obtained from the intercept. The values of yield coefficient (Y) gradually decreased from 0.582 to 0.478 mg of MLVSS/mg of SCOD with the increase in input substrate (SCOD) level from 500 ± 50 to 1000 ± 50 mg/L. This trend indicates a gradual build up of substrate inhibition for heterotrophs under increased SCOD condition which has been corroborated by a consequent increase in the magnitude of endogenous decay rate constant (k_d) from 0.058 to 0.067 for an initial SCOD concentration of 500 ± 50 and 1000 ± 50 mg/L respectively.

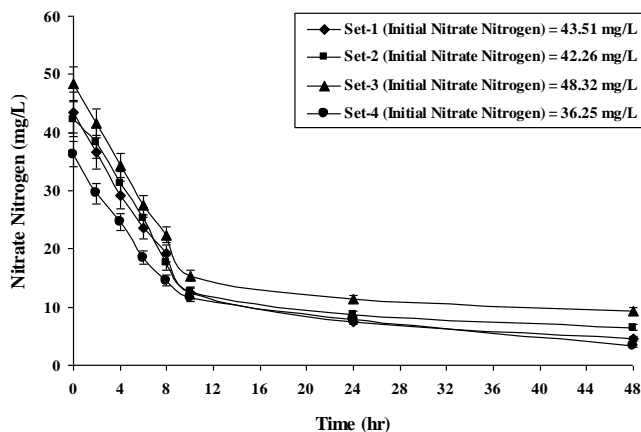


Fig.-7: Nitrate Nitrogen ($\text{NO}_3^-\text{-N}$) Removal Profile during Denitrification Study in Batch Reactor [Initial $\text{NO}_3^-\text{-N}$ Concentration of 40 ± 10 mg/L]

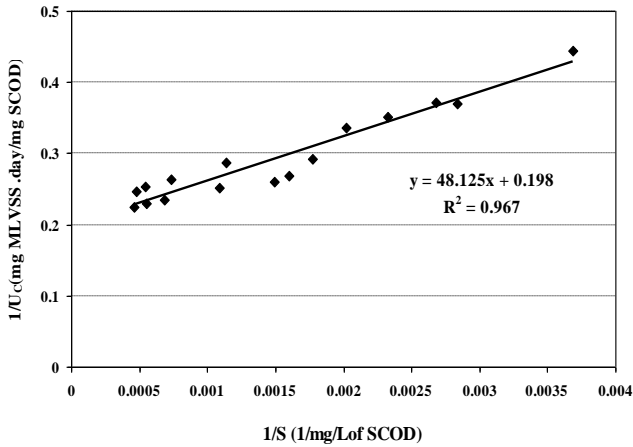


Fig.-9: Substrate Utilization Kinetic for Carbon Oxidation Study in Batch Reactor [Initial SCOD Concentration of 500±50 mg/L]

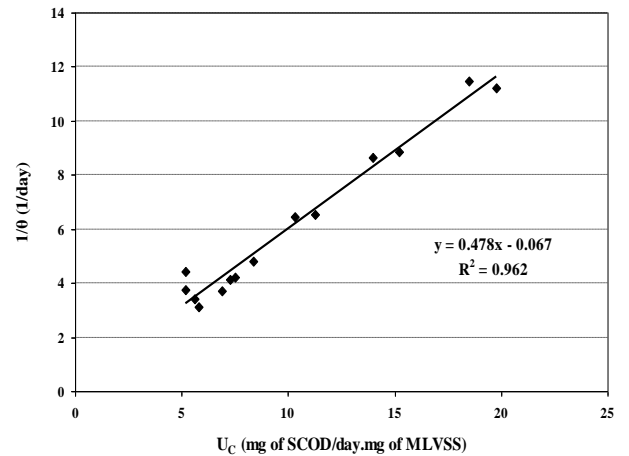


Fig.-12: Microbial Growth Kinetic for Carbon Oxidation Study in Batch Reactor [Initial SCOD Concentration of 1000±50 mg/L]

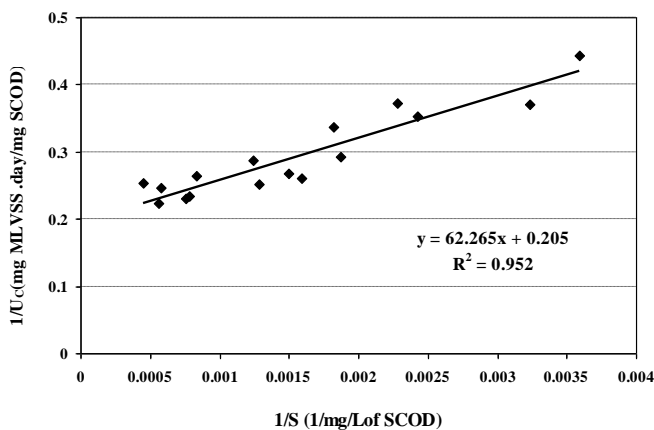


Fig.-10: Substrate Utilization Kinetic for Carbon Oxidation Study in Batch Reactor [Initial SCOD Concentration of 1000±50 mg/L]

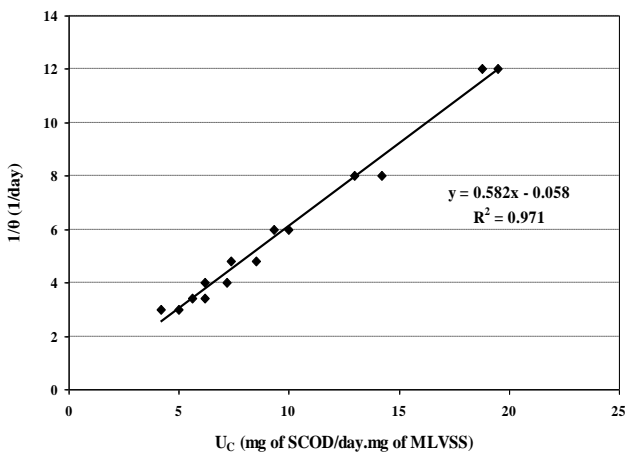


Fig.-11: Microbial Growth Kinetic for Carbon Oxidation Study in Batch Reactor [Initial SCOD Concentration of 500±50 mg/L]

3.5 KINETICS FOR NITRIFICATION

The values for the reciprocal of specific substrate ($\text{NH}_4^+\text{-N}$) utilization rate ($1/U_N$) were plotted against the reciprocal of limiting ammonia nitrogen ($1/N$) and ammonia nitrogen removal kinetics were evaluated using equation $1/U_N = (K_s/k)(1/N) + 1/k$. A best fit graph was drawn by applying a least square method using experimental data which are shown in Fig.-13 and Fig.-14. The slope and intercept of the straight line are K_s/k and $1/k$ respectively. The values of k were 21.27 per day and 17.24 per day for initial $\text{NH}_4^+\text{-N}$ concentration of 90 ± 10 and 140 ± 10 mg/L as N respectively. The maximum substrate utilization rate (k) slightly decreased in case of high initial $\text{NH}_4^+\text{-N}$ concentration of 140 ± 10 mg/L as N indicating a gradual build up of substrate inhibition for autotrophs within the batch reactor. The initial ammonia concentration maintained all along the present study was very high in the tune of 90 to 140 mg/L as N, which is not usually present in any municipal wastewater stream. Consequently, the magnitude of half velocity constant (K_s) value was found to be higher as listed in Table-VIII than the standard values also shown in Table-IX considered for nitrification of municipal wastewater stream [1].

The values of the reciprocal of the reaction time ($1/\theta$) were plotted against specific substrate ($\text{NH}_4^+\text{-N}$) utilization rate (U_N) and shown in Fig.-15 and Fig.-16. The values of yield coefficient (Y) gradually decreased from 0.258 to 0.185 mg of MLVSS/mg of $\text{NH}_4^+\text{-N}$ with the increase in input substrate ($\text{NH}_4^+\text{-N}$) level from 90 ± 10 to 140 ± 10 mg/L as N. It is established that the substrate inhibition on the autotrophs took place at high $\text{NH}_4^+\text{-N}$ value, which is also corroborated by sharp increase (0.059 to 0.068 per day) in endogenous decay rate constant (k_d) in high $\text{NH}_4^+\text{-N}$ value.

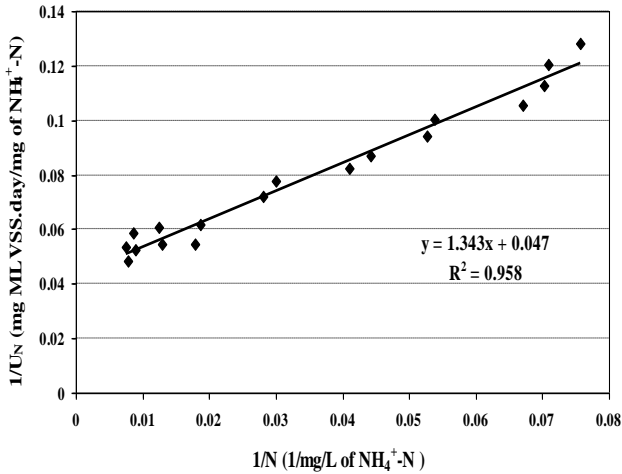


Fig.-13: Substrate Utilization Kinetic for Nitrification Study in Batch Reactor [Initial $\text{NH}_4^+\text{-N}$ Concentration of 90 ± 10 mg/L as N]

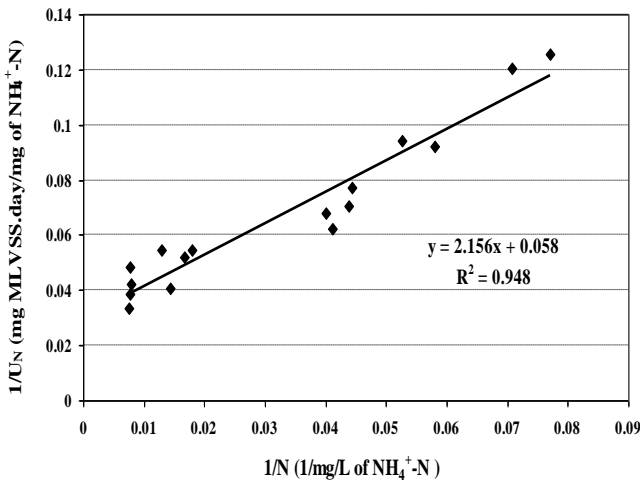


Fig.-14: Substrate Utilization Kinetic for Nitrification Study in Batch Reactor [Initial $\text{NH}_4^+\text{-N}$ Concentration of 140 ± 10 mg/L as N]

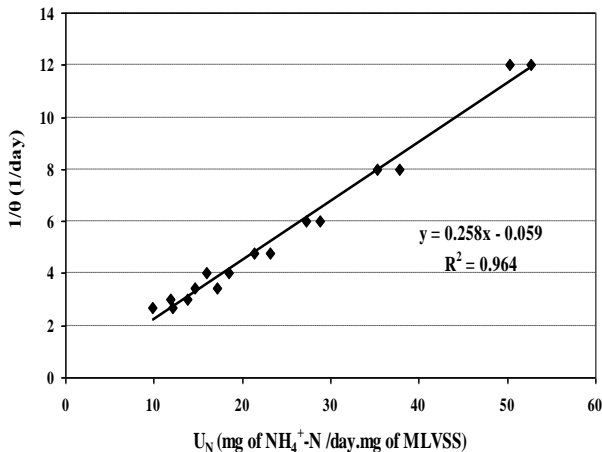


Fig.-15: Microbial Growth Kinetic for Nitrification Study in Batch Reactor [Initial $\text{NH}_4^+\text{-N}$ Concentration of 90 ± 10 mg/L as N]

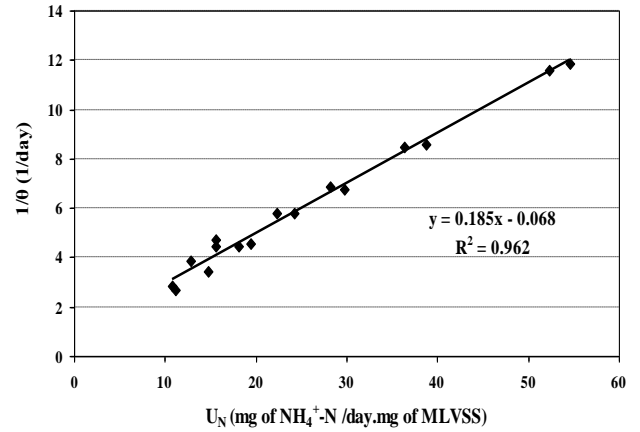


Fig.-16: Microbial Growth Kinetic for Nitrification Study in Batch Reactor [Initial $\text{NH}_4^+\text{-N}$ Concentration of 140 ± 10 mg/L as N]

3.6 KINETICS FOR DENITRIFICATION

Denitrification Kinetics was evaluated separately for initial $\text{NO}_3^-\text{-N}$ concentration of 40 ± 10 and 90 ± 10 mg/L as N as shown in Fig.-17 and Fig.-18. The experimental values for the reciprocal of specific substrate ($\text{NO}_3^-\text{-N}$) utilization rate ($1/U_{\text{DN}}$) were plotted against the reciprocal of limiting nitrate nitrogen ($1/N'$) and nitrate nitrogen removal kinetics were evaluated using equation $1/U_{\text{DN}} = (K_s/k)(1/N') + 1/k$. The slope and intercept of the straight line are K_s/k and $1/k$ respectively. The values of the reciprocal of the reaction time ($1/\theta$) were plotted against specific substrate ($\text{NO}_3^-\text{-N}$) utilization rate (U_{DN}) as shown in Fig.-19 and Fig.-20. The yield coefficient (Y) was determined from the slope of the line and endogenous decay coefficient (K_d) was obtained from the intercept of line. The magnitude of k , Y and k_d as observed from the Table-VIII are in congruence with their respective values that normally be obtained for biological treatment of municipal wastewater as referred in Table-IX. However, the magnitude of K_s (0.287 and 0.357 mg/L for initial $\text{NO}_3^-\text{-N}$ concentration of 40 ± 10 and 90 ± 10 mg/L as N respectively) were obtained higher than that given in Table-IX (0.06 - 0.2 mg/L) perhaps due to high initial $\text{NO}_3^-\text{-N}$ concentration in the reactor.

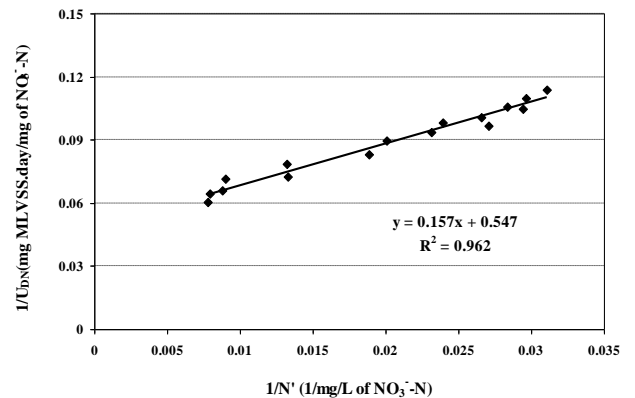


Fig.-17: Substrate Utilization Kinetic for Denitrification Study in Batch Reactor [Initial $\text{NO}_3^-\text{-N}$ Concentration of 40 ± 10 mg/L as N]

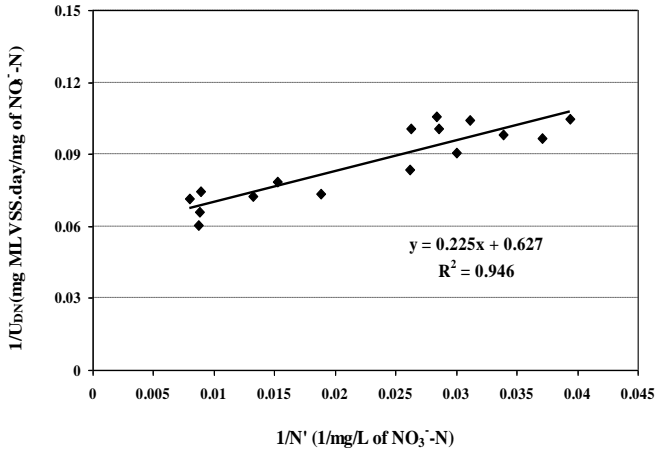


Fig.-18: Substrate Utilization Kinetic for Denitrification Study in Batch Reactor [Initial NO₃⁻-N Concentration of 90±10 mg/L as N]

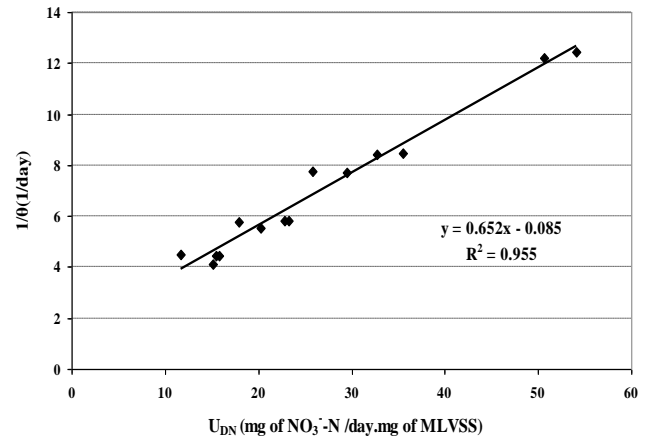


Fig.-20: Microbial Growth Kinetic for Denitrification Study in Batch Reactor [Initial NO₃⁻-N Concentration of 90±10 mg/L as N]

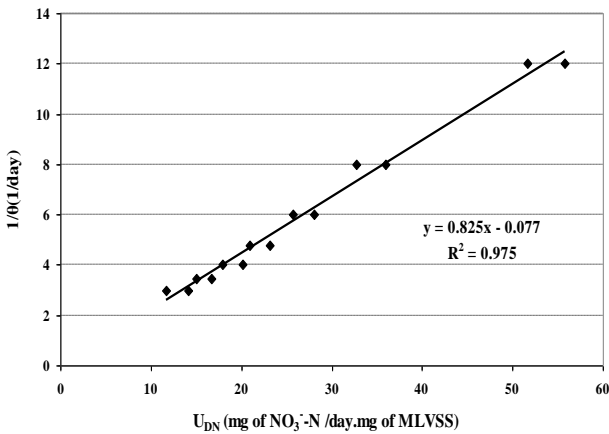


Fig.-19: Microbial Growth Kinetic for Denitrification Study in Batch Reactor [Initial NO₃⁻-N Concentration of 40±10 mg/L as N]

TABLE-VIII: SUMMARIZED VALUES OF BIO-KINETIC COEFFICIENTS FOR BATCH STUDIES

Kinetic coefficients	Carbon oxidation		Nitrification		Denitrification	
	SCOD (500±50 mg/L)	SCOD (1000±50 mg/L)	NH ₄ ⁺ -N (90±10 mg/L)	NH ₄ ⁺ -N (140±10 mg/L)	NO ₃ ⁻ -N (40±10 mg/L)	NO ₃ ⁻ -N (90±10 mg/L)
k (per day)	5.05	4.87	21.27	17.24	1.82	1.59
K _s (mg/L)	243.05	303.23	28.57	37.16	0.287	0.357
Y (mg/mg)	0.582	0.478	0.258	0.185	0.825	0.652
k _d (per day)	0.058	0.067	0.059	0.068	0.077	0.085

TABLE-IX: TYPICAL KINETIC COEFFICIENTS FOR CARBON OXIDATION, NITRIFICATION AND DENITRIFICATION IN MUNICIPAL WASTEWATER [1]

Coefficients	Range for carbon oxidation	Range for nitrification	Range for denitrification
k (per day)	2-10	1-30	0.33-2.25
K _s (mg/L)	25-100	0.2-5.0	0.06-0.2
Y (mg/mg)	0.4-0.8	0.1-0.3	0.4-0.9
k _d (per day)	0.025-0.075	0.03-0.06	0.04-0.08

4. CONCLUSIONS

The mixed microbial culture exhibited a reasonable performance for removal of 86.05-97.39% SCOD and 87.08-93.13% $\text{NH}_4^+\text{-N}$ under batch fed mode. The batch culture kinetics was also performed and kinetic constants were evaluated. The values of bio-kinetic coefficients (k , Y and k_d) for carbon-oxidation and nitrification other than (K_s) were in congruence with their respective typical values reported by some earlier researchers for domestic wastewater treatment. The value of K_s obtained for carbon-oxidation and nitrification were found to be higher than the standard value of kinetics coefficient of municipal wastewater treatment due to higher initial concentrations of SCOD and $\text{NH}_4^+\text{-N}$ in the batch reactor. Denitrification Kinetics were obtained for initial NO_3^- -N concentration of 40 ± 10 and 90 ± 10 mg/L as N separately and the magnitude of k , Y and k_d are observed to be in congruence with their respective values obtained for municipal wastewater. However, the magnitude of K_s (0.287-0.357) was obtained higher than that depicted standard values (0.06-0.2) as initial NO_3^- -N concentration was maintained well above the level usually present in municipal wastewater.

ACKNOWLEDGMENT

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