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Liquid–solid mass transfer in a two phase fluidized bed bioreactor

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Abstract

Liquid–solid mass transfer in a two phase liquid–solid anoxic fluidized bed bioreactor used for biological denitrification has been studied. The denitrification microorganism, immobilized on plastic beads was used as fluidization media. The denitrification rate under different operating conditions was studied and was found to follow zero order kinetics with respect to substrate. The results of experimental work were used to simulate a biofilm model for the evaluation of liquid–solid mass transfer coefficient using the value of concentration obtained at the biofilm surface. The effect of parameters like liquid flow rate and biofilm thickness on mass transfer coefficient were studied. A functional relationship relating Sherwood number to Reynolds and Schmidt number is obtained by regression analysis as $Sh = Re^{0.5985}\ Sc^{0.2048}$.

Keywords: Denitrification; Fluidized bed bioreactor; Biofilm

1. Introduction

Nitrate is the common form of nitrogen in water. It is extensively emitted by industries like fertilizer and explosive industries and is also present in municipal waste. The presence of nitrogen compounds as an inorganic nutrient in the aquatic environment accelerates eutrophication and leads to severe water quality problems especially in lakes. It can also create potential health hazards to human health, because nitrate in the gastrointestinal tract can be reduced to nitrite ions in presence of bacteria. The most common health effects are decreasing the oxygen carrying capacity of blood known as 'Methemoglobinimia' and decreased functioning of thyroid gland. In addition, nitrates and nitrites have the potential to form N-nitrous compounds, which are potent carcinogens.

Though there are a number of biological and physicochemical processes available for removing nitrogen from water and wastewater, in recent years, biological denitrification has drawn attention of many investigators as a powerful technique of wastewater treatment. With this method, organic and ammonia nitrogen are converted into nitrogen gas in anoxic environment. This Nitrogen gas can be safely released to the environment without any risk of pollution. The nitrate reduction reaction involves the following reduction steps [1] in biological method:

$$NO_3^- \rightarrow NO_2^- \rightarrow NO \rightarrow N_2O \rightarrow N_2$$

A wide range of autotrophic as well as heterotrophic bacteria has been shown capable of reducing nitrate to nitrogen gas either in aerobic or anaerobic environment causing denitrfication. They include many in the genera of *Pseudomonas*, *Micrococcus Archobacter*, *Thiobacillus* and *Bacillus*. The most predominant denitrifying bacteria belongs to the genus *Pseudomonas* [2] as most of these bacteria are facultative aerobic organisms with the ability to use oxygen as well as nitrate as final electron donors.

In fluidized bed bioreactors, the particles used as a medium of fluidization can be sand [3,4], expanded clay, activated carbon and charcoal [5] etc. But very little research work has been done using low-density particles as a fluidizing medium. The performance of a fluidized bed reactor can be appropriately estimated by developing a "diffusion-reaction model" over the biofilm in case of attached growth process. This performance evaluation always requires sound calculation of liquid–solid mass transfer resistance to the biofilm and mass transfer resistance within the biofilm. In the literature it has been generally assumed that this liquid–solid resistance can be neglected in the case of higher fluidization rates [6,7]. However, some studies have shown that it influences the biofilm performance significantly, especially at lower fluidization rates and when the bulk substrate concentration is low [8].

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Nomenclature

A	cross sectional area of the column (m^2)
A_p	cross sectional area of a single bioparticle (m^2)
C_S	substrate concentration (kg/m^3)
C_{Sb}	bulk phase substrate concentration (kg/m^3)
C_{Si}	initial nitrate concentration (kg/m^3)
C_{SS}	substrate concentration at biofilm surface (kg/m^3)
C_X	biomass concentration in the reactor (kg/m^3)
d_p	diameter of bioparticle (m)
D	diffusivity of nitrate in biofilm [6] (m^2/s)
h_d	increase in bed height (m)
ID	internal diameter of reactor (m)
k_0	intrinsic zero order rate constant (s^{-1})
K_L	external mass transfer coefficient of nitrate (m/s)
K_S	Monad's constant (kg/m^3)
l	biofilm thickness; $[l = (r_p - r_m) \times 10^6]$ (μm)
n_p	total number of bioparticles
r_c	partial penetration depth (m)
r_m	radius of media particle (m)
r_p	radius of bio particle;$[r_p = [(3/4)Ah_d n_p + r_m{}^3]^{1/3}]$ (m)
r_x	biomass growth rate ($kg/m^3 s$)
$\bar{r}$	characteristic dimension (m)
R_I	intrinsic observed reaction rate ($kg/m^3 s$)
R_{obs}	actual observed reaction rate ($kg/m^3 s$)
$R_{obs,\ model}$	observed reaction rate as predicted by model ($kg/m^3 s$)
Re	Reynolds number; $[Re = (d_p u \rho_w/\mu_w]$
Sc	Schmidt number; $[Sc = (\mu_w/\rho_w D)]$
Sh	Sherwood number $[Sh = (K_L d_p/D)]$
t	time (s)
t_{exp}	time at which exponential growth phase starts (s)
T	time of operation of reactor (s)
u	superficial liquid velocity (m/s)
V	void volume of reactor; $[V = \varepsilon Vo]$ (m^3)
V_o	actual volume of reactor (m^3)
V_s	volume of solids (m^3)

Greek letters

ε	bed voidage $[\varepsilon = 1 - V_s/V_o]$
η	effectiveness factor
μ	specific growth rate of biomass (s^{-1})
μ_{max}	maximum specific growth rate of biomass (s^{-1})
μ_w	viscosity of water (kg/m s)
ρ	biofilm dry density; $[\rho = 104.3 - 0.1245l]$ (kg/m^3)
ρ_w	density of water (kg/m^3)
φ	Thiele modulus
φ_{om}	modified Thiele modulus

In order to study the mass transfer characteristics and effect of liquid–solid mass transfer coefficient on rate of denitrification using low-density particles as fluidizing medium with low nitrate concentrations, experiments have been performed in an anoxic two phase liquid–solid fluidized bed. The effects of initial nitrate concentration and liquid velocities on nitrate removal rate were studied.

2. Materials and methods

2.1. Materials

Experiments were conducted in a tubular glass column (Fig. 1) of 1.5 m height and 75 mm Inner Diameter. Plastic beads with density 1050 kg/m^3 and approximate diameter of 2.89 mm were used as particles for fluidization as well as for the support of microorganism. Substrate broth with composition of 32.6 mg KNO_3, 80.0 mg CH_3OH, 6.0 mg $MgSO_4 \cdot 7H_2O$, 0.2 mg $FeCl_3 \cdot 7H_2O$, 430.0 mg Na_2HPO_4, 320.0 mg NaH_2PO_4/1 l of distilled water [3] was prepared and the pH was adjusted to 7.0 using hydrochloric acid and sodium hydroxide. The broth was then sterilized in an autoclave for 15 min at 15 psi pressure and 120 °C temperature to kill the undesirable microorganisms. Colonies of *Pseudomonas stutzeri* were introduced to the broth after cooling it to room temperature. After inoculating the culture, the broth was kept in an incubator for 24 h where the temperature is maintained at 30 °C at a speed of 60 rpm for better growth of microorganism.

2.2. Analysis methods

Nitrate concentration was estimated using UV Spectrophotometry method [9]. For measuring biomass concentration, 5 ml of sample was collected from the effluent. The initial weight of five empty micro test tubes was noted. Each test tube was filled with 1 ml of sample and centrifuged in a micro centrifuge at 3000 rpm and 25 °C for 15 min so that the biomass will be

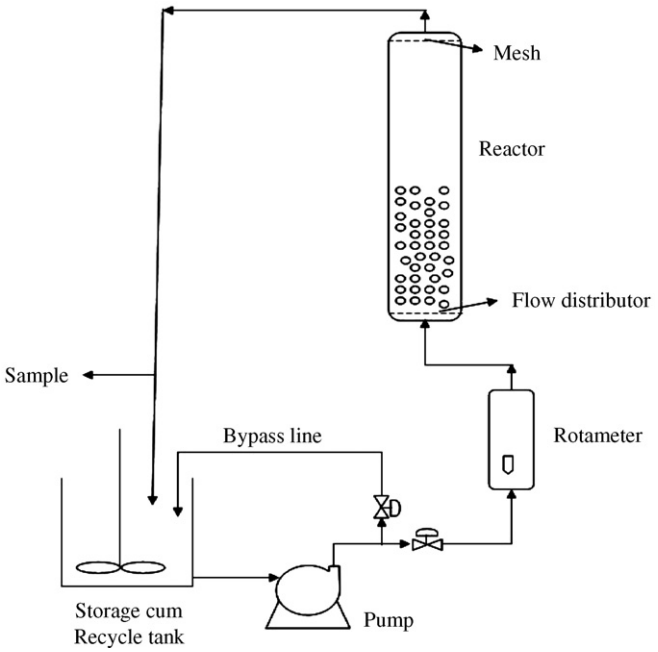

Fig. 1. Experimental setup.

settled at the bottom of tubes. After discarding the supernatant carefully from the tubes, the test tubes were dried in an oven for the removal of moisture and drying of biomass. The final weight of tubes after cooling in desiccators was measured till constant weight is obtained.

- Initial weight of five micro test tubes (mg) = W_1
- Final weight of five micro test tubes (mg) = W_2
- Concentration of biomass (kg/m^3) $C_X = (W_1 - W_2)/5$

2.3. Experimental methodology

Before an experimental run was begun, the bed was seeded with a culture of the denitrifying bacteria so that the microorganism can be immobilized on to the surface of the particles and kept for a night. Synthetic nitrate solution was prepared and added to the recycle cum storage tank. In order to keep the dissolved oxygen low during the reaction, sodium sulfite (100×10^{-3} kg/m^3) was added to the storage tank. Experiments were conducted with initial concentration of nitrate ranging from 10×10^{-3} to 20×10^{-3} kg/m^3, re-circulating liquid flow rate ranging from 4.41 to 5.67 m^3/s. Liquid flow rates were selected between minimum fluidization velocity and velocity corresponding to uniform concentration of solids. Nitrate concentration, pH of the treated water, flow rate and bed height were measured for every 1 h.

3. Results and discussion

3.1. Kinetic parameters estimation

The reaction kinetics in the reactor was studied by conducting experiments with different initial concentration of nitrate. For each initial concentration, the reactor was operated at three different flow rates to study the effect of flow rate. The concentration of nitrate and biomass in the effluent stream at regular intervals was measured.

The biofilm growth rate is first assumed to follow Monad's equation,

$$r_X = \frac{dC_X}{dt} = \mu C_X = \frac{\mu_{max} C_S C_X}{K_S + C_S} \quad (1)$$

After rearrangement and simplification Eq. (1) becomes,

$$\frac{C_S C_X}{r_X} = \frac{C_S}{\mu_{max}} + \frac{K_S}{\mu_{max}} \quad (2)$$

The following procedure was used to evaluate μ_{max} and K_s in Eq. (1)

1. The slope of curve C_X versus t at a point where exponential growth phase starts (t_{exp}) was evaluated which gives the value of r_X.
2. The ordinate term ($C_S C_X/r_X$) was evaluated. Here C_S and C_X corresponds to substrate and biomass concentrations at t_{exp}.
3. The steps 1 and 2 were repeated for various initial nitrate concentrations.

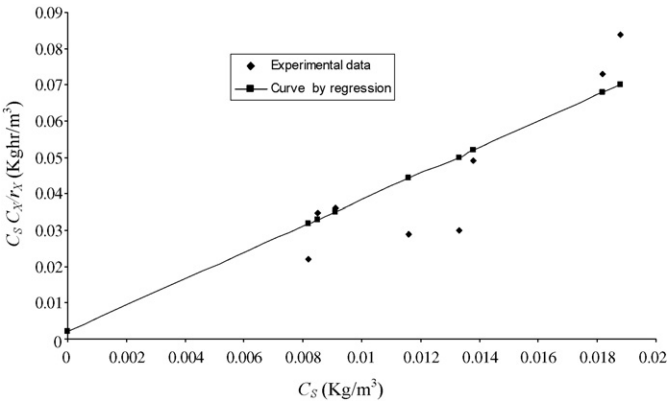

Fig. 2. Estimation of kinetic parameters.

4. Eq. (2) was plotted with C_S versus $C_S C_X/r_X$. The slope gives $1/\mu_{max}$ and intercept gives K_S/μ_{max}. Then the kinetic parameters were evaluated (Fig. 2).

The values obtained are: $\mu_{max} = 0.275$ (h^{-1}) and $K_S = 0.378 \times 10^{-3}$ (kg/m^3).

Since $K_S \ll C_S$, we can say that denitrification reaction, within the experimental results, follows a zero order kinetics with respect to substrate concentration.

3.2. Mass transfer studies

Substrate conversion in a heterogeneous biological fluidized bed reactor can be described by transport of substrate from the bulk liquid phase to the biofilm surface (external mass transfer), transport of the substrate within the biofilm (internal mass transfer) and substrate conversion in the biofilm.

To study the significance of external mass transfer resistance, the substrate consumption rate in the fluidized bed can be related to the overall external mass transfer rate as,

$$R_{obs,model} V = \eta k_0 C_X V = K_L A_P n_P (C_{Sb} - C_{SS}) \quad (3)$$

The mass transfer coefficient (K_L) can be evaluated using the above equation if we know the nitrate concentration at the biofilm surface (C_{SS}). For evaluating nitrate concentration at the biofilm surface, a diffusion reaction model on a spherical bioparticle can be set up using the following assumptions.

1. The bioparticle is spherical in nature and composed of pure culture.
2. The biofilm is regarded as a homogeneous phase within which substrates diffuse and are consumed.
3. Only a single substrate limits the growth (all other nutrients are in excess) and the growth expression is of zero order with respect to substrate concentration.
4. The diffusivity of substrate is assumed to be constant throughout the biofilm and independent of radial position.
5. The effects of outward diffusion of metabolic products are negligible.
6. The limiting substrate is transported in the biofilm via diffusion and obeys first law of Fick's diffusion.

Based on the above considerations, the simultaneous transport and consumption of nitrate within the biofilm can be described by substrate continuity equation:

$$\frac{D}{r^2}\frac{d}{dr}\left(r^2\frac{dC_S}{dr}\right) = R_1 \quad (4)$$

Boundary conditions are:

1. At $r = r_c$, $dC_S/dr = C_S = 0$
2. At $r = r_p$, $C_S = C_{SS}$

By solving the above equation and after applying boundary conditions (Appendix A), we get:

$$C_{SS} = \frac{\rho k_0 r_p^2}{6D} + \frac{\rho k_0 r_c^3}{3Dr_p} - \frac{\rho k_0 r_c^2}{6D} - \frac{\rho k_0 r_c^2}{3D} \quad (5)$$

This can be further modified to get:

$$\frac{{r_c}^3}{{r_p}^3} - 1.5\frac{{r_c}^2}{{r_p}^2} + \frac{1}{2} - \frac{3}{\phi^2} = 0 \quad (6)$$

where $\phi = r_p(\rho k_0/DC_{SS})^{0.5}$

A modified zero order Thiele modulus defined by Mulcahy et al. [6] is given as,

$$\phi_{om} = \bar{r}\left(\frac{\rho k_0}{DC_{SS}}\right)^{0.5} \quad (7)$$

Where $\bar{r} = (4/3\pi({r_p}^3 - {r_m}^3))/4\pi{r_p}^2$

The effectiveness factor for zero order reaction can be given as:

$$\eta = \frac{1-(r_c/r_p)^3}{1-(r_m/r_p)^3} \quad (8)$$

Knowing initial and final concentration of substrate and time of operation of fluidized bed reactor, the observed reaction (i.e., substrate removal rate) is calculated as,

$$R_{obs} = \frac{(C_{Si} - C_{Sb})}{T} \quad (9)$$

The nitrate concentration at the biofilm surface can be calculated as follows:

1. With the values of initial and final concentration and time of operation the actual observed reaction was calculated by Eq. (9).
2. Biofilm thickness was calculated from initial and final bed heights, which intern because of increase in bioparticle volume.
3. Assuming substrate concentration on biofilm surface, Thiele modulus was obtained from Eq. (7).
4. The value of r_c/r_p was obtained from Eq. (6) using the value of Thiele modulus obtained from step 3.
5. From the values of r_c/r_p and r_m/r_p the effectiveness factor was calculated using Eq. (8)
6. The observed reaction rate as predicted by the model is determined from Eq. (3)
7. If the error between $R_{obs,\ model}$ and R_{obs} is within the tolerance limit (10–15%) one can proceed to next step or else has to go back to step 3 and iterate until convergence is achieved.

By knowing all other terms, i.e. A_p, n_p, C_{sb}, K_o, C_X, V and η overall mass transfer coefficient K_L can be calculated.

The data can be fitted to an empirical dimensionless correlation relating Sherwood, Reynolds and Schmidt numbers as

$$Sh = a_0(Re)^b(Sc)^c \quad (10)$$

The coefficients a_0, b and c were obtained by multiple regression analysis. The values obtained are: $a_0 = 1$, $b = 0.5895$, $c = 0.2048$.

Hence the model obtained for calculating external mass transfer coefficient is,

$$Sh = (Re)^{0.5895}(Sc)^{0.2048} \quad (11)$$

3.3. Factors affecting external mass transfer coefficient

3.3.1. Biofilm thickness

The effect of biofilm thickness on external mass transfer coefficient is seen from Table 1. From the table it is clear that, at a particular flow rate the external mass transfer coefficient increases with increasing biofilm thickness and decreases with decrease in biofilm thickness. Hence the change in biofilm thickness brings a similar change in external mass transfer coefficient. This trend may be due to increase in the diameter of bioparticle, which results in the increase of Reynolds number.

Another reason for increasing external mass transfer coefficient may be due to the increase in bed porosity with increase in biofilm thickness. In the literature [10,11], it is reported that biofilm has a heterogeneous internal structure and consisted of

Table 1
Effect of flow rate and biofilm thickness on external mass transfer coefficient

Initial nitrate concentration $\times 10^3$ (kg/m^3)	Flow rate $\times 10^5$ (m^3/s)	Biofilm Thickness $\times 10^4$ (m)	$K_L \times 10^6$ (m/s)
10	4.41	5.983	8.79
	5.04	5.555	6.93
	5.67	5.110	5.91
13	4.41	7.17	9.16
	5.04	6.98	8.57
	5.67	6.59	7.71
15	4.41	7.72	30.93
	5.04	7.356	11.18
	5.67	7.18	7.41
17.5	4.41	5.334	5.978
	5.04	5.110	4.359
	5.67	4.638	3.55
20	4.41	4.876	1.325
	5.04	4.64	1.47
	5.67	4.114	1.62
25	4.41	4.14	1.31
	5.04	3.55	1.21
	5.67	3.12	1.15

channels surrounded by cell clusters. Recently it also has been shown experimentally that internal biofilm structure varies with biofilm thickness. Therefore one can anticipate that increasing number of openings on the biofilm surface for facilitated transfer of substrate is due to increase in biofilm porosity with biofilm thickness.

Another reason for increasing mass transfer coefficient may be due to increase in the roughness of biofilm surface due to increase in biofilm thickness. In the literature [10] it has been also shown that surface roughness influences mass transfer coefficient. Thus increased roughness at higher biofilm thickness values can be also responsible for higher mass transfer coefficient values.

3.3.2. *Effect of initial nitrate concentration on biofilm thickness*

The effect of initial nitrate concentration on mass transfer coefficient is given in Table 1. From the table it reflects that the biofilm thickness increases when the initial nitrate concentration increases and reaches a maximum and decreases afterwards for any particular flow rate. This increasing trend is followed till the initial nitrate concentration is 15 kg/m^3. With further increase in initial nitrate concentration, biofilm concentration reduces continuously and reaches minimum at 25 kg/m^3. This is clearly due to decrease in the activity of microorganisms at higher concentration, which results in reduction of substrate consumption by them, causing biomass concentration to decrease and thus the biofilm thickness.

It is reported in the literature [7] that there exists a biofilm thickness at which substrate conversion is maximum. In the present work also it is noticed that the biofilm thickness is maximum at an initial nitrate concentration of 15 ppm and gave rise to maximum external mass transfer coefficient at this concentration. As substrate conversion increases with increase in biomass, substrate conversion also will be maximum at this initial concentration.

3.3.3. *Liquid flow rate*

The effect of flow rate on mass transfer coefficient can be seen in Table 1. It clearly indicates that, as flow rate increases the value of external mass transfer coefficient decreases, irrespective of initial nitrate concentration. As velocity through column increases, Reynolds number increases. As mass transfer coefficient is proportional to Reynolds number, it should increase. But there also exists an opposite effect of decreasing biofilm thickness with rising velocity. Hence the value of mass transfer coefficient depends upon the parameter that dominates (increase in velocity or decrease in biofilm thickness).

In present experimental work it is seen that effect of decreasing biofilm thickness is dominating. Hence, mass transfer coefficient is reducing with increasing flow rate of liquid.

4. Conclusions

The influence of various parameters like initial concentration of nitrate, flow rate on denitrification rate was studied in the present work. Based on experimental results, analysis of reaction kinetics was done. It was found that biological denitrification follows zero order reaction.

In previous models [5] it was assumed that external mass transfer resistance was low and neglected. In the present study the effect of external resistance was considered. The results of experiments were also used to simulate a biofilm model. During simulation it was found that the biofilm surface concentration of substrate was nearly 60–70% of bulk surface concentration. Hence it can be concluded that external mass transfer resistance was not negligible, and should be considered in the model. An empirical relation for calculation of external mass transfer coefficient was also established. The effect of biofilm thickness and flow rates on K_L was studied. It was found that K_L increases with increasing biofilm thickness and decreases with increasing flow rate.

Appendix A

The simultaneous transport and consumption of nitrate within the biofilm can be described by Eq. (4)

For the partial penetration case, integration of Eq. (4) yields an expression for substrate penetration depth (r_c) as a function of bioparticle radius (r_p) and zero order Thiele modulus which can be derived as follows.

For a zero order reaction, $R_1 = \rho k_0$

Substituting for R_1 in Eq. (4) and rearranging the equation yields,

$$\frac{d}{dr}\left(r^2\frac{dC_S}{dr}\right) = \frac{\rho k_0 r^2}{D} \tag{A1}$$

Integrating Eq. (A1) with respect to r, gives:

$$\frac{dC_S}{dr} = \frac{\rho k_0 r}{3D} + \frac{C_1}{r^2} \tag{A2}$$

Boundary condition 1: at $r = r_c$, $dC_S/dr = C_S = 0$.

Using boundary condition 1, constant C_1 is evaluated and is obtained as,

$$C_1 = -\frac{\rho k_0 {r_c}^3}{3D} \tag{A3}$$

Substituting for C_1 in Eq. (A2) and integrating with respect to r yields,

$$C_S = \frac{\rho k_0 r^2}{6D} + \frac{\rho k_0 r_c^3}{3Dr} + C_2 \tag{A4}$$

Boundary condition 2: at $r = r_c$, $C_S = 0$.

Using boundary condition 2, constant C_2 is evaluated, which is obtained as,

$$C_2 = -\frac{\rho k_0 {r_c}^2}{6D} - \frac{\rho k_0 {r_c}^2}{3D} \tag{A5}$$

Substitution of C_2 in Eq. (A4) gives,

$$C_S = \frac{\rho k_0 r^2}{6D} + \frac{\rho k_0 r_c^3}{3Dr} - \frac{\rho k_0 r_c^2}{6D} - \frac{\rho k_0 r_c^2}{3D} \tag{A6}$$

Boundary condition 3: $C_S = C_{SS}$, at $r = r_p$

Substitution of boundary condition 3 in Eq. (A6) gives,

$$C_{SS} = \frac{\rho k_0 r_p^2}{6D} + \frac{\rho k_0 r_c^3}{3Dr_p} - \frac{\rho k_0 r_c^2}{6D} - \frac{\rho k_0 r_c^2}{3D} \quad \text{(A7)}$$

Simplifying Eq. (A7) yields Eq. (6) mentioned in the manuscript.

$$\frac{{r_c}^3}{{r_p}^3} - 1.5\frac{{r_c}^2}{{r_p}^2} + \frac{1}{2} - \frac{3}{\phi^2} = 0$$

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