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Kinetics of nitrogen removal in high rate anammox upflow filter

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ABSTRACT

The process kinetics for laboratory-scale anammox (anaerobic ammonium oxidation) upflow filter using synthetic wastewater as feed were investigated. The experimental unit consisted of a 2.0 L reactor filled with three-dimensional plastic media. The filter was tested for different influent substrate concentrations and hydraulic retention time (HRT). The substrate loading removal rate was compared with prediction of Stover–Kincannon, second-order and the first-order substrate removal models. Upon approaching pseudo-steady-state condition, substrate ammonium or nitrite concentrations were increased from 280 to 462 mg N/L, while HRT was stepwise decreased from 14.4 to 2 h, with a concomitant increase in nitrogen loading rate (NLR) from 0.93 to 7.34 g/L day. Based on calculations, Stover–Kincannon model and second-order "Grau" model were found to be the appropriate models to describe the upflow filter. According to Stover–Kincannon model, the maximum total substrate removal rate constant (U_{max}) and saturation value constant (K_B) were suggested as 12.4 and 12.0 g N/L day, respectively. As Stover–Kincannon model and second-order model gave high correlation coefficients (97.9% and 98.6%, respectively), these models may be used in predicting the behavior or design of the anammox filter.

1. Introduction

Nitrogenous compounds like ammonium (NH_4^+) are predominant in many wastewaters needing treatment prior to discharge in order to prevent oxygen depletion and eutrophication of surface water bodies. Nitrogen removal is usually accomplished through sequential nitrification and denitrification processes [1,2]. Ammonium (NH_4^+) is oxidized to nitrate (NO_3^-) followed by its reduction to gaseous nitrogen (N_2) during such processes. The anammox process is a novel and promising low cost alternative of conventional nitrogen removal systems to treat nitrogenous compounds [3,4]. Under anoxic conditions, NH_4^+ is oxidized to gaseous N_2 using nitrite (NO_2^-) as electron acceptor with the production of meager amounts of NO_3^- (Eq. (1)) [5] that saves oxygen and organic matter requirements compared with conventional nitrification/denitrification process. The anammox was discovered in Delft, The Netherlands, and it has been observed in many other places [6,7]. Recently, the reaction has been detected in marine sediments and estuarine sediments [8,9]. It is well established that autotrophic bacteria belonging to the order *Planctomycetales* carry out anammox reaction [7,10].

$$NH_4^+ + 1.32NO_2^- + 0.066HCO_3^- + 0.13H^+ = 1.02N_2 + 0.26NO_3^- + 0.066CH_2O_{0.5}N_{0.15} + 2.03H_2O \quad (1)$$

A shortcoming in the full scale application of the anammox process is the requirement of a long start-up time, which may be due to slow growth rates of anammox bacteria (the doubling time was reported to be approximately 11 days) [5,11]. Moreover, the anammox reactor must be efficient in the biomass retention.

The upflow anaerobic filter (AF) has been widely used for the treatment of a variety of wastewaters with different strengths [12]. Young and McCarty [13] demonstrated that it was an effective and feasible technology for wastewater treatment. The AF is basically a contact process in which wastes pass over or through biomass growing on a fixed media contained in the reactor. Therefore, the media acts as a gas–solids separator that helps to provide uniform flow through the reactor, improves contact between the waste constituents and the biomass, and permits accumulation of the large amounts of biomass resulting in longer solids retention time [14]. The AF was also assumed as suitable reactor to carry out anammox [15].

Process modeling is an accepted route for describing the performance of biological treatment systems and predicting their performance. Many models exist in the wastewater treatment literature [16]. First-order substrate removal model [17,18], Stover and Kincannon [18] model and second-order models like Optaken [19] and Grau et al. [20] models are some of the commonly used models to test the kinetics of organic removal in bioreactors. However,

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limited information is available on the process kinetics of substrate removal in anammox filter.

We evaluated AF used for anammox process under different operational conditions like varying HRT and influent substrate concentrations. Different mathematical models, including first-order kinetic model, second-order kinetic model, and Stover–Kincannon model, were applied to the reactor and kinetic coefficients were calculated.

1.1. Theoretical development

1.1.1. First-order substrate removal model

Assuming that first-order kinetics is prevailed in the reactor, the rate of change in substrate concentration in complete mixed system could be expressed as under:

$$-\frac{dS}{dt} = \frac{QS_0}{V} - \frac{QS}{V} - k_1 S \quad (2)$$

Under pseudo-steady-state conditions, the rate of change in substrate concentration ($-dS/dt$) is negligible and the equation given above can be modified as:

$$\frac{S_0 - S}{HRT} = k_1 S \quad (3)$$

where S_0 and S are the influent and effluent substrate concentrations (mg/L); k_1 the first-order substrate removal rate constant (1/d); Q the flow rate of wastewater (L/d) and V is the clean-bed volume of the filter (L).

The value of k_1 can be obtained by plotting $((S_0 - S)/HRT)$ versus S in Eq. (3), which is obtained by rearranging Eq. (2). The value of k_1 can be obtained from the slope of the line.

1.1.2. Stover–Kincannon model

Stover–Kincannon is one of the most widely used mathematical models for determining the kinetic constants in immobilized systems. The model has been applied to continuously operated mesophilic and thermophilic upflow anaerobic filters for the treatment of paper-pulp liquors [14], simulated starch wastewater [21], anaerobic filter for soybean wastewater treatment [12], anaerobic hybrid reactor [22], and anaerobic migrating blanket reactor [23]. However, this model has not been applied for the determination of nitrogen removal kinetic constants in anammox reactor.

Equations of the Stover–Kincannon model are as follows:

$$\frac{dS}{dt} = \frac{Q}{V}(S_0 - S) \quad (4)$$

where dS/dt is defined as follows:

$$\frac{dS}{dt} = \frac{U_{max}(QS_0/V)}{K_B + (QS_0/V)} \quad (5)$$

$$\left(\frac{dS}{dt}\right)^{-1} = \frac{V}{Q(S_0 - S)} = \frac{K_B}{U_{max}}\frac{V}{QS_0} + \frac{1}{U_{max}} \quad (6)$$

where dS/dt, substrate removal rate (g/L day); U_{max}, the maximum utilization rate constant (g/L day) and K_B is the saturation value constant (g/L day).

If $(dS/dt)^{-1}$ is taken as $V/[Q(S_0 - S)]$, which is the inverse of the loading removal rate and this is plotted against the inverse of the total loading rate V/QS_0, a straight line portion of intercept $1/U_{max}$ and a slope of K_B/U_{max} result.

1.1.3. Second-order substrate removal model

The general equation of a second-order model is given below [20]:

$$-\frac{dS}{dt} = k_{2(S)} X \left(\frac{S}{S_0}\right)^2 \quad (7)$$

if Eq. (7) is integrated and then linearilized to get the Eq. (8):

$$\frac{S_0 HRT}{S_0 - S} = HRT + \frac{S_0}{k_{2(S)} X} \quad (8)$$

If the second term of the right part of this equation is accepted as a constant, equation will be modified as under:

$$\frac{S_0 HRT}{S_0 - S} = a + b HRT \quad (8)$$

where $a = S_0/(k_{2(S)}X)$ and b is a constant. $(S_0 - S)/S_0$ expresses the substrate removal efficiency and is symbolized as E. Therefore, Eq. (8) can be written as follows:

$$\frac{HRT}{E} = a + b HRT \quad (9)$$

where X is the average biomass concentration in the reactor (g/L), and $k_{2(S)}$ is the second-order substrate removal rate constant (1/d).

2. Materials and methods

2.1. Synthetic wastewater

Ammonium and nitrite were supplemented to a mineral medium as needed in the form of $(NH_4)_2SO_4$ and $NaNO_2$, respectively. The composition of the mineral medium was (g/L except for trace element solution): $KHCO_3$ 1.25, $NaH_2PO_4{\cdot}2H_2O$ 0.029, $CaCl_2{\cdot}2H_2O$ 0.3, $MgSO_4{\cdot}7H_2O$ 0.2, $FeSO_4$ 0.00625, EDTA 0.00625, and 1.25 mL/L of trace elements solution. The trace element solution contained (g/L) (adapted from van de Graaf et al. [24]): EDTA (15), $ZnSO_4{\cdot}7H_2O$ (0.43), $CoCl_2{\cdot}6H_2O$ (0.24), $MnCl_2{\cdot}4H_2O$ (0.99), $CuSO_4{\cdot}5H_2O$ (0.25), $NaMoO_4{\cdot}2H_2O$ (0.22), $NiCl_2{\cdot}2H_2O$ (0.19), $NaSeO_4{\cdot}10H_2O$ (0.21), H_3BO_4 (0.014), and $NaWO_4{\cdot}2H_2O$ (0.050).

2.2. Inoculum

Activated sludge taken from a mesophilic digester of a municipal wastewater treatment plant served as the inoculum. The seed sludge contained 71.8 g/L suspended solids (SS) and 46.1 g/L of volatile suspended solids (VSS).

2.3. Bioreactor

A simplified flow-sheet of the reactor is shown in Fig. 1a. The reactor was composed of plexiglass with an effective volume of 2 L, an internal diameter of 70 mm and effective height of 520 mm. The filter was operated in continuous mode and it was packed with string shaped three-dimensional plastic media (Yixing, China) to retain biomass as illustrated in Fig. 1b. This string consisted of bundles of the soft fibrous media which were evenly spaced at intervals of 30 mm, affixed at the center-line of the column. Its specific surface area was about 400 m^2/m^3. The filter was covered with black cloth to avoid the inhibition caused by light and was operated at $30 \pm 1\,^{\circ}C$ and its pH was maintained in range of 7.5–8.0.

2.4. Start-up and operation

The start-up was accomplished by inoculating the reactor with sludge containing 30 g VSS/L. At 30 °C, initially the HRT was set at 24 h with the influent substrate (NO_2^-–N or NH_4^+–N) concentration of 70 mg/L, corresponding to nitrogen loading rate (NLR) of 0.14 g/L day. The NLR was increased stepwise by raising substrate concentrations. Within 3 months, the influent substrate concentration was increased to 280 mg/L, with a concomitant increase in NLR to 0.56 mg/L day.

After a satisfactory start-up, the reactor was operated until a pseudo-steady-state was reached which was indicated by a

Table 1
Performance of anammox reactor at different influent substrate concentrations during Phase I.

Influent concentration (mg/L)		Effluent concentration (mg/L)			Substrate removal efficiency (%)			Loading rate (g N/L day)	
NH_4^+–N	NO_2^-–N	NH_4^+–N	NO_2^-–N	NO_3^-–N	NH_4^+–N	NO_2^-–N	Total substrate	Total substrate	Total substrate removal
280	280	7.1	4.1	59.5	97.5	98.5	98.0	0.933	0.915
308	308	4.1	6.9	72.1	98.7	97.8	98.2	1.03	1.01
336	336	4.7	13.3	77.6	98.6	96.1	97.3	1.12	1.09
364	364	2.3	22.8	79.4	99.4	93.7	96.6	1.21	1.17
420	420	32.3	18.9	72.3	92.3	95.5	93.9	1.40	1.32
448	448	42.3	21.7	59.5	90.6	95.2	92.9	1.49	1.39
462	420	51.8	41.2	72.9	88.8	90.2	89.5	1.47	1.32

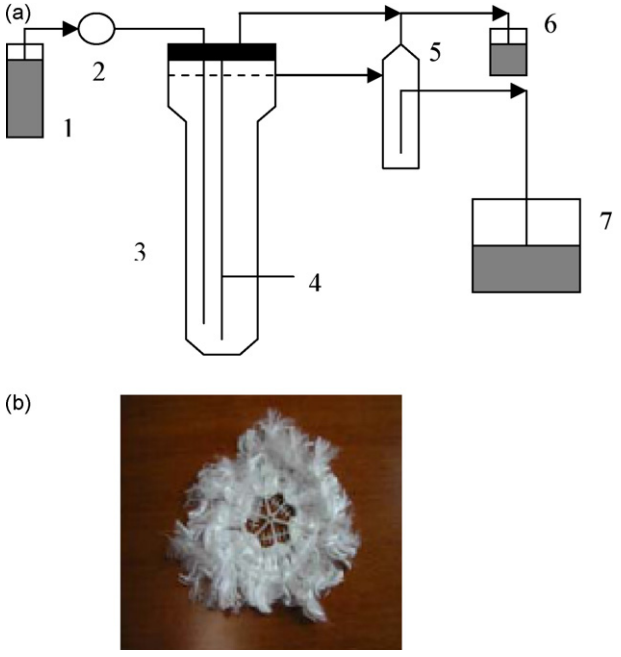

Fig. 1. (a) Schematic diagram of anammox upflow filter system (1) feeding tank, (2) peristaltic pump, (3) reactor, (4) media, (5) gas–liquid–solid separator, (6) gas outlet, and (7) effluent collection tank; (b) image of the packing media.

constant effluent nitrogen concentration (±5%). Two series of experiments were conducted to study the performance of the reactor after the start-up. In Phase I, the substrate concentration was increased from 280 to 462 mg N/L, keeping HRT at 14.4 h. In Phase II, the HRT was decreased gradually from 14.4 to 2 h while keeping the substrate concentration at 280 mg N/L. Each run lasted over 10 days before proceeding to the next condition. Data based on arithmetic means of three or more measurements obtained at pseudo-steady-state were reported.

2.5. Analytical methods

The influent and effluent samples were collected on daily basis and were analyzed immediately or stored in a refrigerator at 4 °C until the analyses were carried out. The measurements of NH_4^+–N, NO_2^-–N, NO_3^-–N, pH, SS and VSS were performed according to the Standard methods [25].

3. Results and discussion

3.1. Reactor performance

The performance of the reactor under different influent substrate concentrations and HRTs are shown in Tables 1 and 2. Results indicated that very high substrate removal efficiencies were attained even at NLR as high as 7.3 g/L. The NLRs in the present study are comparable to those obtained during previous researches, which are summarized in Table 3. Literature survey indicated that NLR values in a range of 1.0–8.9 g/L day could be achieved in anammox reactors, and the NLRs were quite high compared to those in conventional nitrification/denitrification systems i.e. 0.3–0.5 g/L day [35].

3.2. First-order substrate removal model

The value of k_1 was obtained from the slope of the line by plotting $(S_0 - S)$/HRT versus S in Eq. (2). Fig. 2 shows that the correlation coefficient of the plot was 0.172. The low value of the coefficient (R^2) clearly indicates that first-order kinetics cannot be applied with fair degree of precision.

Table 2
Performance of anammox reactor at different operating HRTs during Phase II.

HRT (h)	Influent concentration (mg/L)		Effluent concentration (mg/L)			Substrate removal efficiency (%)			Loading rate (g N/L day)	
	NH_4^+–N	NO_2^-–N	NH_4^+–N	NO_2^-–N	NO_3^-–N	NH_4^+–N	NO_2^-–N	Total substrate	Total substrate	Total substrate removal
10.1	270	288	28.7	5.8	69.6	89.4	98.0	93.8	1.32	1.24
8.33	270	288	67.5	28.6	77.3	75.0	90.1	82.8	1.61	1.33
6.82	270	288	79.5	31.8	67.0	70.6	89.0	80.1	1.96	1.57
6.00	297	307	92.4	44.6	67.0	68.9	85.5	77.3	2.42	1.87
5.52	295	304	111	48.0	60.4	62.3	84.2	73.4	2.60	1.91
4.68	295	304	114	60.6	66.7	61.4	80.1	70.9	3.07	2.18
4.08	305	304	115	68.6	62.5	62.3	77.4	69.9	3.58	2.50
3.94	305	304	111	63.2	65.1	63.7	79.2	71.5	3.71	2.65
3.60	305	304	114	70.4	60.4	62.5	76.8	69.6	4.06	2.83
3.05	305	304	120	70.4	60.9	60.6	76.8	68.7	4.80	3.29
2.69	305	304	94.4	41.8	60.2	69.1	86.3	77.6	5.44	4.22
2.40	305	304	78.1	17.6	68.9	74.4	94.2	84.3	6.09	5.13
2.21	305	304	96.3	15.2	63.7	68.4	95.0	81.7	6.62	5.41
1.99	305	304	78.6	23.2	61.1	74.2	92.4	83.3	7.34	6.11

Table 3
Resume of the operation of different anammox reactors.

Reactor	Inlet	Support material	NLR (g/L day)	Reference
Fixed bed	Synthetic medium	Glass beads	1.1	[27]
Fixed bed	Sludge dewatering effluent	Soft media	0.6	[28]
Fluidized bed	Synthetic medium	Sand	1.8	[27]
Fluidized bed	Sludge digester effluent	Sand	1.5	[27]
Membrane bioreactor	Synthetic medium	–	0.7	[29]
Granular sludge bed	Synthetic medium	–	2.1	[30]
UASB	Synthetic medium	–	2.5	[31]
SBR	Synthetic medium	–	1.0	[5]
SBR	SHARON effluent	–	2.4	[32]
SBR	Fish canning effluent	–	0.7	[33]
Gas-lift	Synthetic medium	–	8.9	[26]
SBR	Synthetic medium	–	2.7	[34]
Filter	Synthetic medium	Three-dimensional-plastic media	2.5	[34]
Filter	Synthetic medium	Three-dimensional-plastic media	7.3	Present work

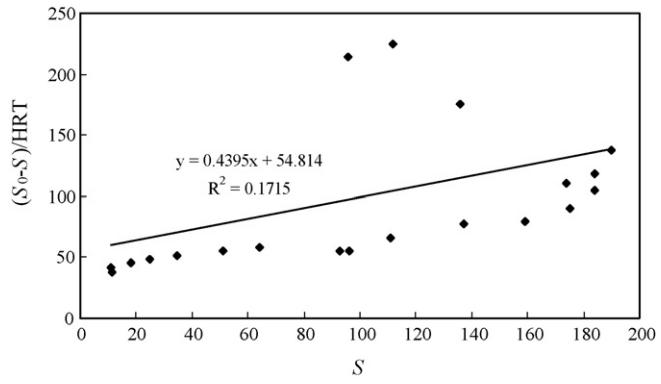

Fig. 2. First-order kinetics model plot. S is the total substrate (sum of ammonium–nitrogen and nitrite–nitrogen) concentrations.

3.3. Stover–Kincannon model

Fig. 3 shows the graph plotted between reciprocal of total substrate removal rate, $V/[Q(S_0 - S)]$, against the reciprocal of NLRs, $V/(QS_0)$. According to Eq. (5), saturation constant (K_B) value and maximum total substrate utilization rate (U_{max}) were calculated from the line plotted on graph in Fig. 3 as 12.0 and 12.4 g N/L day. Experimental data containing high correlation ($R^2 = 0.979$) was applied to the model. Table 2 showed that the maximum total substrate removal rate of the reactor was 6.1 g N/L day, merely 49.2% of U_{max}, suggesting that the reactor possessed an excellent nitrogen removal capacity even better than the highest reported value (Table 3).

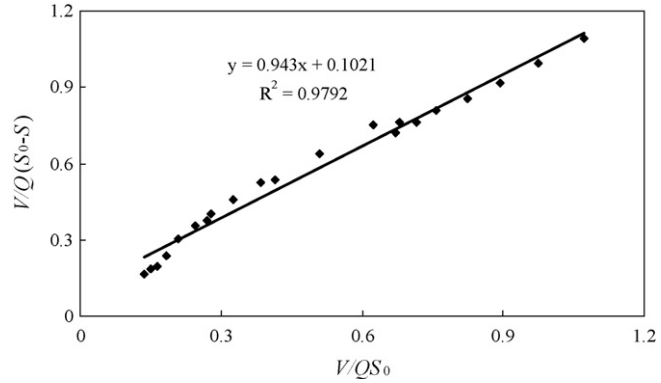

Fig. 3. Stover–Kincannon model plot.

Table 4
Data for second-order kinetics model for anammox filter.

HRT (h)	S_0 (mg/L)	S (mg/L)	E (%)	HRT/E (h)
14.4	560	11.2	98	14.69
14.4	616	11.0	98.2	14.66
14.4	672	18.0	97.3	14.80
14.4	728	25.1	96.6	14.91
14.4	840	51.2	93.9	15.34
14.4	896	64.0	92.9	15.50
14.4	882	93.0	89.5	16.09
10.1	558	34.5	93.8	10.77
8.33	558	96.1	82.8	10.06
6.82	558	111	80.1	8.51
6.00	604	137	77.3	7.76
5.52	599	159	73.4	7.52
4.68	599	175	70.9	6.60
4.08	609	184	69.9	5.84
3.94	609	174	71.5	5.51
3.60	609	184	69.6	5.17
3.05	609	190	68.7	4.44
2.69	609	136	77.6	3.47
2.40	609	95.7	84.3	2.85
2.21	609	112	81.7	2.71
1.99	609	102	83.3	2.39

From substrate mass balance of the influent and effluent volumes, following equation can be obtained:

$$S = S_0 - \frac{12.6S_0}{12.0 + (QS_0/V)} \quad (10)$$

The model developed for prediction of total substrate removal efficiency in the present work is as follows:

$$E = \frac{12.6}{12.0 + (QS_0/V)} \quad (11)$$

3.4. Second-order substrate removal model

Data used for second-order kinetic model are given in Table 4. Using Fig. 4 for the filter (a) and (b) values were obtained which were 1.397 and 0.964, respectively, with correlation coefficient of 0.986. The formula for predicting effluent substrate concentration for the filter is given by;

$$S = S_0\left(1 - \frac{\mathrm{HRT}}{1.397 + 0.964\mathrm{HRT}}\right) \quad (12)$$

while substrate removal efficiency is represented by

$$E = \frac{\mathrm{HRT}}{1.397 + 0.964\mathrm{HRT}} \quad (13)$$

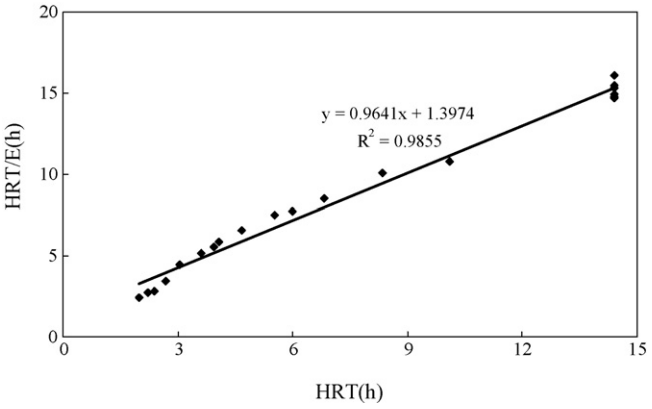

Fig. 4. Second-order kinetics model plot.

4. Conclusion

Treatment performance of the anammox filter was evaluated at different substrate concentrations, HRTs and NLRs using synthetic wastewater and kinetic analyses of the reactor were carried out according to the experimental results. Upon reaching pseudo-steady-state, substrate concentration was increased from 280 to 462 mg/L by decreasing HRT stepwise from 14.4 to 2 h, with a concomitant increase in NLR from 0.93 to 7.34 g/L day. Substrate removal efficiencies ranged from 71.5% to 98.2% during the experimental studies.

Biokinetic models such as first-order, second-order and Stover–Kincannon models were applied for the anammox filter. Second-order model and Stover–Kincannon model gave higher correlation coefficients of 98.6% and 97.9%, respectively. Therefore, these models may be used in the design of the anammox filter.

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