Impact of seeding with nitrifying bacteria on nitrification process efficiency

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Abstract Seeding of nitrifying bacteria into the activated sludge process was studied both theoretically and experimentally. A simple model was developed for prediction of the effects of seeding of nitrifying bacteria from a separate stage into the activated sludge process. The purpose of seeding is to improve the treatment results and the process stability as well as to decrease the volume requirements of the process. Pilot plant studies were carried out at the Uppsala municipal wastewater treatment plant in order to evaluate the effects of seeding. One line was supplied with supernatant from dewatering of digested sludge and the nitrification process gave an activated sludge with a high fraction of nitrifying bacteria, suitable for seeding. The other line was supplied with pre-precipitated wastewater and with the excess sludge from the line treating the supernatant. The experimental results showed that nitrification could be obtained at sludge ages that would otherwise preclude nitrification. Performance relationships for the system developed, based on laboratory and on-line measurements were studied and are presented. The studies show that seeding may decrease the necessary volume needs for a stable nitrification process and that the effects could be predicted by use of a simple model.
Keywords Activated sludge; bioaugmentation; digested supernatant; modelling; nitrification; pilot plant; seeding

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Introduction

Many different terms can be used for description of the process of addition of specialised bacteria to the activated sludge process: seeding, bioaugmentation, bacterial augmentation, biomass enhancement and inoculum addition. The purpose of seeding is to improve the nitrogen removal process efficiency, nitrification process stability and to decrease the volume requirements of the process.

A decrease of nitrification efficiency has often been observed at Swedish wastewater treatment plants during the snow thawing period with high flows and low temperatures (Plaza *et al.*, 1988). In order to obtain a high efficiency of nitrification during winter time at low temperatures, the reactor volume need may be even three times higher in the winter period than in the summer period. Seeding technology is one of the methods for reduction of the process volume requirements through decreasing the critical sludge age. Excess activated sludge with a high fraction of nitrifying bacteria may be seeded into another activated sludge tank to facilitate the nitrification process.

The effects of seeding nitrifying bacteria into the activated sludge process have been studied based on theoretical models (Finnson, 1994; Lee, 1997; Li and Hultman, 1997; Rittman, 1996 and 1997; and Tendaj-Xavier, 1985) and experimental works (Daigger *et al.*, 1993; Parker and Richards, 1994; Rittman and Whiteman, 1994; and Sinkjaer *et al.*, 1996).

Theoretical background

A system with seeding of nitrification bacteria is shown in Figure 1. The system consists of one line for treatment of supernatant from dewatering of digested sludge and one line for treatment of wastewater from the primary clarifiers. In the figure some notations used in models are shown.

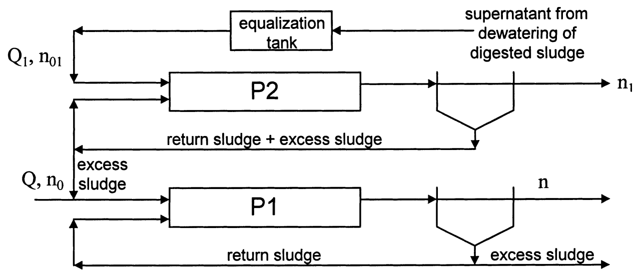

Figure 1 System with seeding of nitrification bacteria

The line for supernatant is used for production of nitrification bacteria. The produced amount may be written:

$$X_0 = Y_n (n_{01} - n_1) Q_1 \tag{1}$$

where

X_0 =produced amount of nitrification bacteria

Y_n =yield coefficient for nitrification bacteria

n_{01} =influent ammonium concentration to the supernatant line

n_1 =effluent ammonium concentration from the supernatant line

Q_1 =influent flow to the supernatant line.

The influent concentration of nitrification bacteria, X_{0N}, to the wastewater treatment line is:

$$X_{0N} = Y_n (n_{01} - n_1) \frac{Q_1}{Q + Q_e} \tag{2}$$

where

Q =influent flow to the wastewater treatment line

Q_e =excess sludge flow from aeration basin of the supernatant line

The measured sludge age, Θ_{daer}, calculated for the aerated zone of the activated sludge process is defined as:

$$\Theta_{daer} = \frac{A}{B + N} \tag{3}$$

where

A =sludge mass in the aeration zone

B =removed mass of sludge per day from the activated sludge process without seeding

N =removed additional mass of sludge per day from the activated sludge process due to supply of seeding sludge

In seeding technology of the activated sludge process it is necessary to consider both the net growth rate of the specific bacteria and the measured sludge age. The net growth rate of nitrifying bacteria, μ_N, may be written as (Rittman, 1996):

$$\mu_N = \frac{M_N - QX_{0N}}{X_N V_{aer}} \tag{4}$$

where

M_N =amount of nitrifying bacteria per day from the activated sludge process in the excess sludge and in the effluent

Q =influent flow rate

X_N=concentration of nitrifying bacteria in the aerated zone of the activated sludge process

V_{aer} =volume of the aerated zone.

The fraction of nitrifying bacteria is assumed to be the same in the aerated zone as in the sludge from the activated sludge process (*B*+*N*). In this case the removed nitrogen concentration for an activated sludge process with complete mixing and stationary conditions may be written (Li and Hultman, 1997):

$$n_0 - n = \frac{X_{0N}}{Y_n}\frac{\mu_N \Theta_{daer}}{1-\mu_N \Theta_{daer}} \tag{5}$$

where

n =effluent ammonium concentration

n_o =influent ammonium concentration that can be nitrified.

For lower ammonium concentrations the Monod relationship may be applied:

$$\mu_N = \frac{\mu_{Nmax} n}{K_n + n} \tag{6}$$

where

K_n =half saturation coefficient

μ_{Nmax} =maximum growth rate of nitrifying bacteria.

If formula (6) is inserted in formula (5) the following equation is obtained:

$$n_0 - n = \frac{X_{0N}}{Y_n}\frac{\Theta_{daer}}{\frac{K_n + n}{\mu_{N\,max} n} - \Theta_{daer}} \tag{7}$$

which may be arranged as:

$$n^2 - n\left(n_0 - \frac{K_n + \frac{X_{0N}\mu_{N\,max} n \Theta_{daer}}{Y_n}}{1-\mu_{N\,max}\Theta_{daer}} \right) - \frac{K_n n_0}{1-\mu_{N\,max}\Theta_{daer}} = 0 \tag{8}$$

and gives the solution:

$$n = \frac{a}{2} \pm \sqrt{\frac{a^2-4b}{4}}\, a = n_0 - \frac{K_n + \frac{X_{0N}\mu_{N\,max} n \Theta_{daer}}{Y_n}}{1-\mu_{N\,max}\Theta_{daer}}\, b = -\frac{K_n n_0}{1-\mu_{N\,max}\Theta_{daer}} \tag{9}$$

The positive sign before the square root in formula (9) is used when $\Theta_{daer}<1/\mu_{Nmax}$ and the negative sign when $\Theta_{daer}>1/\mu_{Nmax}$.

Simulation of effluent values of ammonium (n) from the wastewater line may be done by use of formula 9 for different values of μ_{Nmax}, K_n, X_{0N}/Y_n and Θ_{daer}. Experimental determinations of different parameters may be done as follows:

- X_{0N}/Y_n by measurements of influent and effluent ammonium concentrations of the supernatant line and the flows Q, Q_1 and Q_e
- μ_N by use of formula 5 and measurements of aerobic sludge age (formula 3) and of influent and effluent ammonium concentrations of the wastewater line
- μ_{Nmax} and K_n by use of formula 6 for different determined values of μ_N and *n*.

In order to compensate for temperature effects the following temperature dependence may be used:

where $\mu_{N,15}$=nitrification growth rate at 15°C, and T=temperature, °C

Materials and methods

Pilot plant description. The pilot plant consisted of two separate lines with the activated

$$\mu_{N,15} = \mu_N * 1.127^{(T-15)} \qquad (10)$$

sludge process, suitable for testing various operation strategies and modes. The volume of the activated sludge tank in each line was 2.35 m^3 and the settling tanks had a volume of 0.55 m^3 each. The pilot plant was equipped with various on-line meters (Figure 2) and with a specially designed novel computerized control supervision system (CCS). It included the monitoring and control of flows and oxygen concentration in the nitrification tank. The on-line measurements data (DO, ammonia, nitrate, suspended solids, pH, redox and temperature) were collected and stored by the computer system and were evaluated by Matlab, Excel and a special program written in Turbo Basic (Carlsson *et al.*, 1997).

Experimental strategy. In the studied system earlier presented in Figure 1, pre-sedimentated wastewater, pumped from the full-scale plant to the pilot plant influent tank, was continuously supplied to Line 1 (P1). Line 2 (P2) was fed with supernatant from dewatering of digested sludge. The supernatant water was pumped to P2 from a storage tank which was placed outdoors. Sodium hydrogen carbonate was added as alkalinity source to keep the pH value at about 7. The consumption of sodium hydrogen carbonate was about 4.2 kg per 1 kg of ammonia nitrogen oxidized.

The experiment was performed in three steps. During the first step, a steady stage full nitrification of supernatant was obtained for Line 2. During the second step, the excess sludge from Line 2 with high fractions of nitrifying bacteria was seeded into Line 1. Meanwhile Line 1 was operated at a sludge age under the critical value. During the third step, the dynamic effect of seeding was investigated.

Sampling procedure and analyses. Throughout the whole experiment 24-h composite samples were collected by vacuum samplers from the influent and effluent lines twice a week. For both lines the following analyses were performed: COD, filter COD, Tot-N, NH_4–N, (NO_2+NO_3)–N, HCO_3 and SS. Periodically an intensive sampling program was introduced with 24-h composite samples taken every day. Analytical procedures were performed according to the Swedish standards (SIS).

Batch tests. Potential nitrification rates were determined indirectly by oxygen utilization rate (OUR) tests. The activated sludge taken from the pilot plant was placed in a water bath with a temperature of 20°C and was continuously aerated (usually to a level of 6 to 8 mg

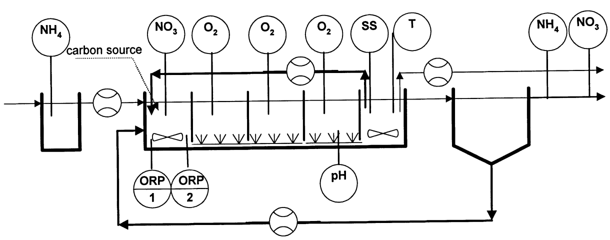

Figure 2 Layout of the pilot-scale activated sludge plant

Table 1 Characteristics of the supernatant

Parameter	Average concentration, g/m^3	St. dev.
COD (total)	650	325
Tot-N	343	122
NH_4–N	290	60
Tot-P	15.5	3.6
PO4–P	10	2.2
SS	345	88.6
HCO_3	880	8.9*

*after addition of bicarbonate

Table 2 Summary of operational data

Parameter	Range
Aerobic sludge age, days	5–8.6
Temperature, ºC: –5/4–24/4	6–11.6
–25/4–11/5	6–13.1
–12/5–8/3	11–18.6
Reject water flow, l/h	50–100
MLSS, g/l	2–5
SVI, ml/g	66

O_2/l). In order to determine the nitrification rat two measurements of respiration rates were carried out. Oxygen utilization rates for activated sludge with ammonia and with allyl-thioura (ATU) were measured by an oxygen probe which was introduced into a 250 ml BOD bottle. The OUR values were calculated from the resulting oxygen utilization profiles. The net value of OUR calculated from the OUR value for activated sludge with ammonia minus the OUR value for activated sludge with ATU was considered to be contributed to nitrification. Using a conversion factor of 4.33 g O_2/g N a potential nitrification rate was estimated.

Results

Experiments with nitrification of supernatant

Operational performance. Experiments with nitrification of supernatant from dewatering of digested sludge were carried out in Line 2 of the pilot plant. The characteristics of the supernatant are presented in Table 1. The experiments started at an aerobic sludge age of about 6.7 days as suggested by the simulation. During the first five weeks it was difficult to get a stable nitrification process since a high SS concentration in the effluent from Line 2 was observed and the actual aerobic sludge age was only about five days. The temperature of the supernatant water was often lower than 10ºC. In order to get a higher nitrification efficiency the aerobic sludge age was increased to about eight days by reducing excess sludge flow rate to 5 1/h beginning from the 20th of April. An effect of substantial decreasing of ammonia concentration in the effluent by increasing of sludge age can be observed in Figure 3. The average value of the nitrification efficiency obtained in Line 2 was 88%.

Operational results for the supernatant treatment plant are summarized in Table 2. The influent flow varied from 50 l/h to 100 l/h. The pilot plant was operated with the return sludge flow of 220 l/h and an internal recirculation rate of 660 l/h. An excess sludge in an amount of 5 l/h was pumped directly from the last zone of the activated sludge tank. During

Table 3 Characteristics of the pre-precipitated wastewater

Parameter	Concentration, average, g/m^3	St. dev.
COD	190	86.3
CODf	100	26.5
Tot-N	26.3	7.2
NH_4–N	20	3.7
NO_2+NO_3–N	0.35	0.39
Tot-P	3	1.5
PO_4–P	0.76	0.53
SS	80	24.9
HCO_3	400	37.4

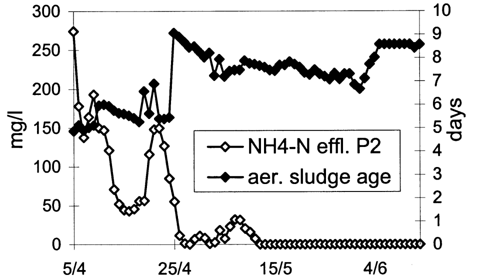

Figure 3 Ammonia nitrogen concentrations in the effluent (on-line) and the aerobic sludge age

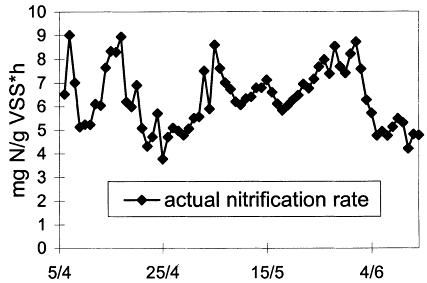

Figure 4 Actual nitrification rate – Line 2

the seeding operation mode, the excess sludge was pumped into Line 1.

Nitrification process. The variability in actual nitrification rate is presented in Figure 4. The mean value of 6.8 mg N/g VSS * h was obtained for the period analysed. No inhibition effect of the influent on the nitrifying bacteria was observed during this period. The potential nitrification rate was measured during the period between April and June. The potential nitrification rate was about 8.3 mg N/g VSS * h for the sludge taken from Line 2. Laboratory tests showed that the ratio between nitrification rates of the sludge from Line 2 and Line 1 was about 3 when complete nitrification was obtained in Line 2. The fraction of nitrifying bacteria in the sludge treating the municipal wastewater is about 3%. Using the factor 3, as obtained from tests, the fraction of nitrifying bacteria in the investigated sludge was estimated to about 9%.

Experiments with seeding of nitrifying bacteria

Operational performance. The nitrifying bacteria produced in Line 2 were seeded to an activated sludge process fed with pre-precipitated wastewater (Line 1). Characteristics of the pre-precipitated wastewater are presented in Table 3.

The seeding of the activated sludge from Line 2 to Line 1 started on May 15. In the seeding experiments, nitrifying bacteria were obtained from the pilot plant line with nitrification of supernatant from dewatering of digested sludge. All excess sludge from this line was supplied to the line where the seeding effects were studied except for 3 July–10 July, when seeding was stopped. Line 1 was operated at the flow rate of 220 l/h, which corresponds to the hydraulic retention time in the aeration tank of 11 h. The return sludge flow was 220 l/h and the internal recirculation rate 660 l/h. The excess sludge flow was 20 l/h (15/5–23/5), 40 l/h (24/5–6/6), 60 l/h (7/6–20/7) and 40 l/h (21/7–3/8) with the corresponding average aerobic sludge ages of 2.6, 1.3, 0.8 and 1.3 days, respectively. Aeration basin temperatures ranged from 11.3 to 17.9ºC and averaged 14.6ºC.

Figure 5 shows the time series of the measured aerobic sludge age during the time of study. By comparison of these values with the ammonia concentrations in the effluent, it can

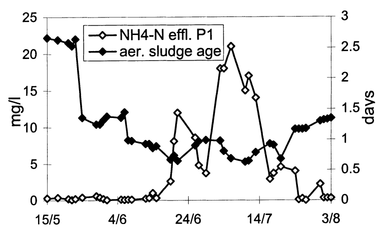

Figure 5 Aerobic sludge age and effluent ammonium concentrations (no seeding 2–10 July)

be observed that nitrification can be maintained at lower aerobic sludge age than would be possible if seeding did not occur. It can be seen that a high removal efficiency could be obtained for nitrification in the seeded line for sludge ages down to about 0.5 days at a temperature of 17ºC. There was no seeding of nitrification bacteria between 3–10 July which explains the high peak value of ammonium during that period. The experiments showed that nitrifying bacteria from the line for treatment of supernatant retained their activity after they had been transferred to the line treating pre-precipitated wastewater. The very large effect of seeding of sludge with nitrifying bacteria on the required sludge age depends on the large amount of seeded sludge. In practice a much lower amount of seeded sludge can be used.

During the experiments with seeding of nitrification bacteria the temperature increased from about 11ºC to 18ºC. Temperature effects on the growth of nitrification bacteria were compensated by use of formula 10. Different methods are available to determine coefficients in the Monod relationship (formula 6) (Roberts, 1977). Best correlation was obtained by the Hanes method, while other methods showed much less correlation (Figure 6). Obtained values for the coefficients $\mu_{N,max,15}$ and Kn were 0.11 d^{-1} and 0.016 g N/m^3, respectively. Hanes method, however, gives emphasis on higher values of the ammonium concentration. In comparison of experimental values of the effluent ammonium concentration, *n*, with calculated values of *n* by use of formula 9, a reasonably good agreement was obtained after 19/6 (Figure 7). For the first period the values of $\mu_{N,max,15}$, had to be increased to 0.20 d^{-1} in order to obtain agreement between measured and calculated values.

Discussion

Seeding of sludge from a separate nitrification stage may be an advantageous technology in implementing nitrogen removal at anew plants or in upgrading of existing plants. Many studies have shown that ammonia rich supernatant may be oxidized by nitrifying bacteria to nitrate in a separate nitrification step (Mossakowska *et al.*, 1997 and Tendaj-Xavier, 1985). It can be operated in a reliable way at temperatures around 20ºC throughout the whole winter.

A very high dosage of nitrification bacteria was used in the studies (X_{0N}/Y_n=135 g N/m^3 as an average value) and for this reason those bacteria dominated compared with the produced bacteria in the wastewater line. The high dosage was used to document the effects of seeding. The quotient of flow of supernatant and of pre-precipitated wastewater (Q_1/Q) was 0.45 during the seeding experiments and thereby much higher than in existing full-scale plants. Two different ways may be used to increase the seeding effects in existing plants. The first is to seed nitrification bacteria to only one or two lines in a plant with several lines. Excess sludge from these lines may be added to the other lines. The second way is to increase the ammonium content in the supernatant, for instance by special conditioning methods of sludge (i.e. heat treatment or addition of acids or bases) or addition of nitrogen rich wastes (as septic sludge and urine) directly to the sludge handling stream. Possibilities may also be considered to store nitrifying sludge or nitrogen rich streams for use during crucial conditions for nitrification in the main stream.

The model described in this paper shows that it is possible to calculate in a simple way the effects of seeding. An important assumption is that the seeded nitrifying bacterium will maintain its activity in the activated sludge processes. Activity losses due to changed environmental conditions and grazing of protozoa on the bacteria may reduce the positive seeding effects.

The line treating supernatant could be operated in a stable way at about five days at 15ºC. The maximum nitrification growth rate was therefore above 0.2 d^{-1}. A good agreement between simulated and experimental data was obtained in the wastewater treatment line for $\mu_{N,max,15}$=0.20 d^{-1} and 0.11 d^{-1} for the first month and the following period of the seeding experiments, respectively. This means that some change in environmental conditions caused a decrease of the maximum nitrification growth rate.

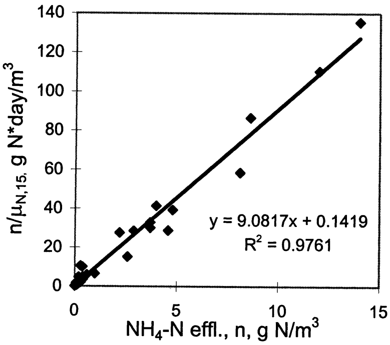

Figure 6 Determination of parameters in Monod model

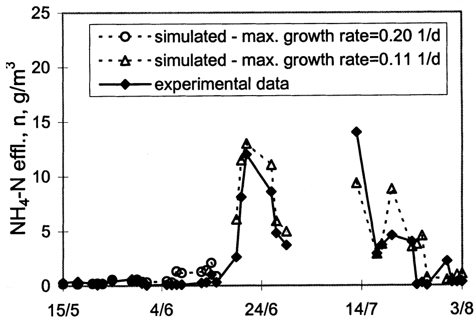

Figure 7 Verification of model with experimental data during seeding conditions: K_n=0.016 g N/m^3

Conclusions

- Seeding of nitrifiers from a separate step for treatment of supernatant (Line 2) into an activated sludge process treating pre-precipitated wastewater (Line 1) made it possible to maintain a stable nitrification process in Line 1 at low sludge ages which would otherwise preclude nitrification.
- The minimum value of the sludge age that may be used for the treatment of supernatant in order to secure a stable and efficient nitrification process at 15°C was five days.
- A simple model was developed for prediction of the effects of seeding of nitrifying bacteria from a separate stage into the activated sludge process and the model gave a reasonably good agreement between calculated and experimentally obtained results.
- The maximum growth rate of nitrification bacteria at 15°C was about 0.2 d^{-1} during the first month of the seeding experiments and diminished to about 0.11 d^{-1} during the next period.

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