Operating conditions analysis for a partial nitrification process with biomass retention.*

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Abstract: In wastewater treatment, ammonium removal is a key step which can be done biologically. One method is by coupling a partial nitrification with the Anammox process. The partial nitrification goal is to convert half of the ammonium into nitrite, so ammonium and nitrite can be later converted into dinitrogen gas by Anammox bacteria. To obtain a stable partial nitrification, ammonium oxidizing bacteria (AOB) have to prevail over nitrite oxidizing bacteria (NOB) so as to avoid further conversion of nitrite into nitrate. Two control objectives can be identified for partial nitrification: the repression of NOB and the regulation of the effluent to obtain the required nitrite to ammonium ratio. In this work, the equilibrium points of the partial nitrification process are analyzed to find operating conditions for AOB prevalence over NOB. Based on this analysis, a feeding strategy is proposed to regulate the effluent nitrite to ammonium ratio at the value required by Anammox. The study is based on the process dynamical model in a reactor with biomass retention.

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1. INTRODUCTION

Ammonia removal in wastewater via anaerobic ammonium oxidation (Anammox) is one of the current alternatives to conventional nitrification-denitrification methods. Some of its main advantages are the reduced costs in aeration and lack of requirement of external carbon sources, since the process is fully autotrophic (Ali and Okabe, 2015). Anammox bacteria can convert ammonium into dinitrogen gas with nitrite as electron donor under anoxic conditions (Guo et al., 2013). The required nitrite can be obtained from partial nitrification (van Dongen et al., 2001; van Loosdrecht and Salem, 2006; Wu et al., 2016).

Nitrification is a two step reaction in which ammonium is converted into nitrite (nitrification) by ammonium oxidizing bacteria (AOB), which is later converted into nitrate (nitratation) by nitrite oxidizing bacteria (NOB). Partial nitrification consists in only oxidating the ammonium while the oxidation of nitrite is prevented (van Dongen et al., 2001; Volcke, 2006; Isanta et al., 2015). As nitratation is unwanted and reduces the process efficiency; AOB must prevail over NOB in the interspecies competition. The unwanted bacterial group must be repressed through the selection of adequate operation parameters.

Studying the stability of the equilibrium points of the process aids to obtain analytically such operation parameters. For instance, which flow rates or ammonium concentrations make stable an equilibrium in which NOB goes to zero. The first studies of the equilibrium points of nitrification processes were presented in Volcke et al. (2007, 2010). These results are extended here to processes with biomass retention, as it is commonly encountered in practice. Biomass retention involves different dilution rates for bacteria and dissolved substances.

A second objective of this work is to obtain an appropriate effluent composition to avoid leaving unconverted ammonium or nitrite after Anammox (Guo et al., 2013; van Dongen et al., 2001; van Loosdrecht and Salem, 2006). This objective can also be approached by studying the equilibrium concentrations to choose appropriate operating parameters.

In this work the different equilibrium points of the nitrification process with biomass retention are studied to obtain restrictions on the operating parameters. These restrictions are later used to ensure the survival of AOB over NOB. Having assured NOB repression, flow rates and influent concentrations are defined to obtain the desired effluent composition in views to coupling with Anammox.

2. PROCESS MODEL

We will assume that the nitritation and nitratation processes take place in a reactor operated in continuous mode with biomass retention mechanism. The biomass retention allows setting the bacteria retention time (inverse of dilution rate) different to the hydraulic retention time. Then, the process dynamical model is (Volcke et al., 2007):

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\[ \dot{X}_{\text{AOB}} = (\mu_1 - D_A)X_{\text{AOB}} \]
\[ \dot{X}_{\text{NOB}} = (\mu_2 - D_N)X_{\text{NOB}} \]
\[ \dot{S}_{\text{NH}_3} = D(S^f_{\text{NH}_3} - S_{\text{NH}_3}) - k_{11}\mu_1X_{\text{AOB}} - k_{12}\mu_2X_{\text{NOB}} \]
\[ \dot{S}_{\text{NO}_2} = D(S^f_{\text{NO}_2} - S_{\text{NO}_2}) + k_{21}\mu_1X_{\text{AOB}} - k_{22}\mu_2X_{\text{NOB}} \]

where \( X_{\text{AOB}} \) and \( X_{\text{NOB}} \) are AOB and NOB concentrations, \( S_{\text{NH}_3} \) and \( S_{\text{NO}_2} \) are ammonium and nitrite concentrations in the reactor, and \( S^f_{\text{NH}_3} \) and \( S^f_{\text{NO}_2} \) are the concentrations in the influent stream. \( D \) is the (liquid) dilution rate defined as the influent flow rate \( (F) \) over the reactor volume \( (V) \), and \( D_A \) and \( D_N \) are the dilution rates of AOB and NOB, defined as the inverses of the biomass retention times. Although normally AOB and NOB have the same dilution rate, to make this study more general, they are considered different. If there is no retention mechanism, all the dilution rates are the same: \( D = D_A = D_N \). The \( k_{ij} \) are yield coefficients.

Parameters \( \mu_1 \) and \( \mu_2 \) are the specific growth rates for AOB and NOB, respectively, modeled by Monod kinetics:

\[ \mu_1(S_{\text{NH}_3}) = \frac{\mu_1^m S_{\text{NH}_3}}{k_{\text{NH}_3} + S_{\text{NH}_3}} \]
\[ \mu_2(S_{\text{NO}_2}) = \begin{cases} \frac{\mu_2^m S_{\text{NO}_2}}{k_{\text{NO}_2} + S_{\text{NO}_2}} & \text{if } S_{\text{NH}_3} \neq 0 \\ 0 & \text{if } S_{\text{NH}_3} = 0. \end{cases} \]

Parameters \( \mu_1^m \) and \( \mu_2^m \) are the maximum growth rates, and \( k_{\text{NH}_3} \) and \( k_{\text{NO}_2} \) are affinity constants.

3. EQUILIBRIUM POINTS

To obtain restrictions on the operating parameters we analyze the feasibility and stability of each equilibrium point of the process. The goal is to find the operating conditions that make the only stable equilibrium points those where NOB’s concentration is zero.

The equilibrium points are obtained by setting all the derivatives to zero in (1) and solving the resulting algebraic system, which gives place to four solutions. For each of these equilibria to be realistic, ammonium, nitrite, AOB and NOB concentrations must be non-negative. Moreover, as ammonium is only consumed, its concentration must be lower than the influent one. It is not the case of nitrite which is produced by AOB. To verify that an equilibrium point is feasible the equilibrium concentrations must fulfill:

\[ [S^*_{\text{NH}_3}; S^*_{\text{NO}_2}; X^*_{\text{AOB}}; X^*_{\text{NOB}}] \in \mathbb{R}^4_+ \]

\[ S^*_{\text{NH}_3} \leq S^f_{\text{NH}_3}, \quad S^*_{\text{NO}_2} \leq \min \{S^f_{\text{NO}_2}, S^*_{\text{NH}_3} \} \]

In the following subsections, each of the possible equilibrium points and its feasibility are analyzed.

3.1 Wash-out equilibrium point

The wash-out equilibrium point is that in which all biomass is lost and ammonium and nitrite concentrations go to the input ones:

\[ P_w = \left( S^f_{\text{NH}_3}; S^f_{\text{NO}_2}; 0; 0 \right). \]

This point is always feasible, no matter the dilution rates or influent concentrations. Hence, care must be taken not to fall into it.

3.2 Only AOB survival equilibrium point

The AOB survival equilibrium point is that in which NOB does not survive while AOB does. This is the desired equilibrium point of the process. The equilibrium concentrations are:

\[ P_a = \begin{pmatrix} S^f_{\text{NH}_3} + S^f_{\text{NO}_2} - S^*_{\text{NH}_3} \\ S^f_{\text{NO}_2} - S^*_{\text{NO}_2} \\ k_{11}S^f_{\text{NH}_3} - k_{12}S^f_{\text{NO}_2} \end{pmatrix} \]

\[ P_a = \left( S^f_{\text{NO}_2} + S^f_{\text{NH}_3} - S^*_{\text{NH}_3} \right) \frac{1}{k_{11}S^f_{\text{NH}_3} - k_{12}S^f_{\text{NO}_2}} \]

\[ \text{where } S^*_{\text{NH}_3} \text{ is defined as } \]
\[ S^*_{\text{NH}_3} = \frac{D_A k_{\text{NH}_3}}{\mu_1^m - D_A} \]

\[ \text{for ease of notation.} \]

It is found that the following restriction guarantees the feasibility (3) of the AOB survival equilibrium:

\[ S^f_{\text{NH}_3} > S^*_{\text{NH}_3} \]

which is equivalent to

\[ D_A < \mu_1(S^f_{\text{NH}_3}) \]

That is, for the feasibility of AOB survival over NOB, the influent ammonium being greater than the equilibrium concentration suffices. In terms of dilutions, the limit to AOB’s dilution rate is the maximum achievable growth rate, which is obtained with the highest possible ammonium.

3.3 Only NOB survival equilibrium point

Although unlikely to happen in practice, an equilibrium where NOB prevails over AOB is possible. The equilibrium concentrations are:

\[ P_n = \begin{pmatrix} S^f_{\text{NH}_3} - k_{21} S^f_{\text{NO}_2} - S^*_{\text{NO}_2} \\ S^f_{\text{NO}_2} - S^*_{\text{NO}_2} \\ S^f_{\text{NO}_2} - S^*_{\text{NO}_2} \end{pmatrix} \]

\[ \text{where } S^*_{\text{NO}_2} \text{ is defined as} \]
\[ S^*_{\text{NO}_2} = \frac{D_N k_{\text{NO}_2}}{\mu_2^m - D_N} \]

for ease of notation.

The following restriction guarantees the feasibility (3) of the AOB survival equilibrium:

\[ S^f_{\text{NO}_2} > S^*_{\text{NO}_2} \]
\[ S^f_{\text{NO}_2} < S^*_{\text{NO}_2} + \frac{k_{22}}{k_{12}} S^f_{\text{NH}_3} \]

which are equivalent to

\[ D_N < \mu_2(S^f_{\text{NO}_2}) \]
\[ D_N > \mu_2(S^f_{\text{NO}_2} - \frac{k_{12}}{k_{22}} S^f_{\text{NH}_3}) \]

That is, for the feasibility of NOB survival over AOB, the influent nitrite needs to be inside an interval which
depends on the current influent ammonium. In terms of dilutions, there is an interval defined by the NOB dilution rate and the influent ammonium and nitrite.

3.4 AOB and NOB survival equilibrium point

The last equilibrium is that where both bacterial groups survive. The equilibrium concentrations are

\[
P_x = \left( \begin{array}{c}
\frac{S_{NH3}^A}{S_{NH3}^e} \\
\frac{S_N^A}{S_N^e}
\end{array} \right)
\]

The feasibility of this equilibrium point is given by a set of bounds on ammonium and nitrite:

\[
S_{NH3}^f > S_{NH3}^* + \frac{k_{12}}{k_{22}}(S_{NO2}^f - S_{NO2}^*)
\]

\[
S_{NH3}^f > S_{NH3}^* - \frac{k_{11}}{k_{21}}(S_{NO2}^f - S_{NO2}^*)
\]

\[
S_{NO2}^f > S_{NO2}^* + \frac{k_{21}}{k_{11}}(S_{NH3}^f - S_{NH3}^*)
\]

\[
S_{NO2}^f < S_{NO2}^* + \frac{k_{22}}{k_{12}}(S_{NH3}^f - S_{NH3}^*)
\]

which are equivalent to

\[
D_A < \mu_1(S_{NH3}^f - \frac{k_{12}}{k_{22}}(S_{NO2}^f - S_{NO2}^*))
\]

\[
D_A < \mu_1(S_{NH3}^f + \frac{k_{11}}{k_{21}}(S_{NO2}^f - S_{NO2}^*))
\]

\[
D_N > \mu_2(S_{NO2}^f - \frac{k_{22}}{k_{12}}(S_{NH3}^f - S_{NH3}^*))
\]

\[
D_N < \mu_2(S_{NO2}^f + \frac{k_{21}}{k_{11}}(S_{NH3}^f - S_{NH3}^*))
\]

This is, influent ammonium is lower bounded by some ammonium value greater than the dilution rate and the influent ammonium and nitrite concentrations. The situation is similar for dilutions: there is an upper bound for AOB dilution rate, lower than the one required in the AOB survival point, and there is an interval for NOB dilution rate.

4. STABILITY OF THE EQUILIBRIUM POINTS

Having analyzed the feasibility of each equilibrium point, the next step is to inspect their stability. Local asymptotic stability (Khalil, 2002) is analyzed by determining the location of the eigenvalues of the system linearized at each equilibrium point. This analysis gives place to additional restrictions to complement those for feasibility.

4.1 Wash-out equilibrium point

The eigenvalues of (1) linearized at the wash-out equilibrium point \( P_w \) are:

\[
\lambda_1 = -D
\]

\[
\lambda_2 = \mu_1(S_{NH3}^f) - D_A
\]

\[
\lambda_3 = \mu_2(S_{NO2}^f) - D_N
\]

Clearly the first two eigenvalues are negative for any positive dilution rate. The other two eigenvalues are negative if the following restrictions hold:

\[
D_A > \mu_1(S_{NH3}^f)
\]

\[
D_N > \mu_2(S_{NO2}^f)
\]

which are equivalent to

\[
S_{NH3}^f < S_{NH3}^*
\]

\[
S_{NO2}^f < S_{NO2}^*
\]

It can be observed that the restrictions for stability of the wash-out (17) and (18) oppose to the restrictions for feasibility of the only AOB survival point (7) and (8), and only NOB survival point (11) and (12). Hence, even if the wash-out is always feasible, it will not be stable if the restrictions for the feasibility of any of those two equilibria hold.

Note also that for the wash-out equilibrium stability, a higher bound is imposed over ammonium (18a). This restriction is incompatible with the feasibility conditions of the AOB and NOB survival equilibrium (14a) and (14b), which together set a lower bound for ammonium at a value greater than the one imposed by (18a). Therefore, the wash-out and the AOB and NOB survival equilibrium cannot be stable for the same operating conditions.

4.2 AOB survival equilibrium point

The eigenvalues of (1) linearized at the AOB survival equilibrium point \( P_a \) are:

\[
\lambda_1 = -D
\]

\[
\lambda_2 = \mu_2(S_{NO2}^eq) - D_N - a\mu_1(S_{NH3}^eq)X_{AOB}^{eq} + \frac{1}{2}\sqrt{(D + a\mu_1(S_{NH3}^eq)X_{AOB}^{eq})^2 - 4a\mu_1(S_{NH3}^eq)DAX_{AOB}^{eq}}
\]

where \( \mu_1(S_{NH3}^eq) \) is the partial derivative of \( \mu_1 \) with respect to \( X_{AOB} \) evaluated at \( S_{NH3}^* \):

\[
\mu_1(S_{NH3}^eq) = \frac{\partial\mu_1}{\partial X_{AOB}}|_{S_{NH3}^eq=S_{NH3}^*} = \frac{\mu_1^2 k_{NH3}}{(k_{NH3} + S_{NH3}^*)^2} > 0.
\]

The first eigenvalue is negative for any positive dilution rate. The second eigenvalue is negative if

\[
D_N > \mu_2(S_{NO2}^f - \frac{k_{11}}{k_{21}}(S_{NH3}^f - S_{NH3}^*))
\]

which is equivalent to

\[
S_{NH3}^f > S_{NH3}^* - \frac{k_{11}}{k_{21}}(S_{NO2}^f - S_{NO2}^*)
\]

\[
S_{NO2}^f > S_{NO2}^* - \frac{k_{21}}{k_{11}}(S_{NH3}^f - S_{NH3}^*).
\]

The last two eigenvalues have negative real part as long as the equilibrium point is feasible, that is (7) or (8). This is concluded after comparing the first term with the one with the square root. Moreover, it can be shown that the eigenvalues are real by analyzing the radicand of the second term. For that, it must be accounted that the bacteria dilution rate cannot be greater than the liquid dilution \( D_A \leq D \).
Note at this point that the restrictions (21) and (22) for the stability of this equilibrium point oppose to the ones for the feasibility of the AOB and NOB survival point \( P_2: \) (14b), (14c) and (15d). Hence, if the process is operated to make the AOB survival point feasible and stable, the AOB and NOB survival point is unfeasible, and conversely.

Moreover, the stability restriction (21) for AOB survival stability is incompatible with the feasibility restriction (12) for NOB survival. For this, recall that \( S_{\text{NH}_3}^f < S_{\text{NH}_3}^* \) in the AOB survival equilibrium. Thus, these two equilibria cannot happen under the same operating conditions.

At this point, we can see that the desired equilibrium, AOB survival, can be guaranteed if its feasibility and stability restrictions are met.

### 4.3 NOB survival equilibrium point

The eigenvalues of (1) linearized at the NOB survival equilibrium point \( P_n \) are:

\[
\lambda_1^2 = -D \tag{23a}
\]

\[
\lambda_n^2 = \mu_1(S_{\text{NH}_3}^\text{eq}) - D_A \tag{23b}
\]

\[
\lambda_n^{3,4} = -\frac{D + d\mu_2(S_{\text{NO}_2}^\text{eq})X_{\text{NO}_2}^\text{eq}}{2} + \sqrt{\left(\frac{D + d\mu_2(S_{\text{NO}_2}^\text{eq})X_{\text{NO}_2}^\text{eq}}{2}\right)^2 - 4d\mu_2(S_{\text{NO}_2}^\text{eq})D_N X_{\text{NO}_2}^\text{eq}} \tag{23c}
\]

where \( \mu_2(S_{\text{NO}_2}^\text{eq}) \) is the partial derivative of \( \mu_2 \) with respect to \( X_{\text{NO}_2}^\text{eq} \) evaluated at \( S_{\text{NO}_2}^* \):

\[
\left. \frac{\partial \mu_2}{\partial X_{\text{NO}_2}} \right|_{S_{\text{NO}_2}^*} = \frac{\mu_2^2' k_{\text{NO}_2}}{(k_{\text{NO}_2} + S_{\text{NO}_2}^*)^2} > 0. \tag{24}
\]

The algebraic structure of these eigenvalues is very similar to the ones of the AOB survival equilibrium. So, the analysis is omitted.

The restrictions for the stability of the NOB survival are that the equilibrium point is feasible and

\[
D_A < \mu_1(S_{\text{NH}_3}^f - \frac{k_{12}}{k_{22}}(S_{\text{NO}_2}^f - S_{\text{NO}_2}^*)) \tag{25}
\]

which is equivalent to

\[
S_{\text{NH}_3}^f > S_{\text{NH}_3}^* + \frac{k_{12}}{k_{22}}(S_{\text{NO}_2}^f - S_{\text{NO}_2}^*) \tag{26a}
\]

\[
S_{\text{NO}_2}^f < S_{\text{NO}_2}^* + \frac{k_{22}}{k_{12}}(S_{\text{NH}_3}^f - S_{\text{NH}_3}^*) \tag{26b}
\]

Similarly to the AOB survival equilibrium, in this case the stability conditions (25) and (26) oppose to the feasibility conditions of the AOB and NOB survival equilibrium \( P_2: \) (14a), (14d) and (15c). Hence, if the process is operated to make NOB survival feasible and stable, the AOB and NOB survival is unfeasible, and conversely.

### 4.4 AOB and NOB survival equilibrium point

The eigenvalues at this equilibrium point are harder to compute without numerical substitutions. Hence, an alternative approach has been taken. Due to its length, only an outline of the proof is given here, but it can be examined in detail in Appendix A.

After linearizing (1) at \( P_2 \) the Jacobian matrix is obtained. The sign and trace of the Jacobian are the sum and product of its eigenvalues, respectively. Based on their signs it can be concluded that there are either one or two pairs of eigenvalues with negative real part if the equilibrium point is feasible. Later, the Routh-Hurwitz theorem can be applied. Again, if the equilibrium point is feasible, by noting that there are no sign changes in the Routh tabulation we can conclude that there are no eigenvalues with positive real part.

From this analysis it can be concluded that the same restrictions that apply for this equilibrium point feasibility also apply for its local asymptotic stability, that is, (14) and (15).

### 5. PROCESS OPERATION FOR PARTIAL NITRIFICATION

Throughout the previous section we have shown that the restrictions for feasibility and stability of each equilibrium point oppose to those of the other equilibria. Consequently, only one equilibrium point can be feasible and stable for a set of operating parameters. This is an important result since our goal is to operate only in the AOB survival equilibrium point, which will be possible if its corresponding restrictions are followed. Unfortunately, it is not possible to formally state that this equilibrium is globally stable without proving that no limit cycles can exist, which is not covered in this work.

The conditions for AOB survival and NOB repression are:

\[
S_{\text{NH}_3}^f > S_{\text{NH}_3}^* \tag{27a}
\]

\[
S_{\text{NH}_3}^f < S_{\text{NH}_3}^* - \frac{k_{11}}{k_{21}}(S_{\text{NO}_2}^f - S_{\text{NO}_2}^*) \tag{27b}
\]

\[
S_{\text{NO}_2}^f < S_{\text{NO}_2}^* - \frac{k_{21}}{k_{11}}(S_{\text{NH}_3}^f - S_{\text{NH}_3}^*) \tag{27c}
\]

or equivalently

\[
D_A < \mu_1(S_{\text{NH}_3}^f) \tag{28a}
\]

\[
D_N > \mu_2(S_{\text{NO}_2}^f + \frac{k_{22}}{k_{12}}(S_{\text{NH}_3}^f - S_{\text{NH}_3}^*)) \tag{28b}
\]

A secondary objective is to obtain a given nitrite to ammonium ratio in the process output

\[
\frac{S_{\text{NO}_2}^f}{S_{\text{NH}_3}^f} = \delta \tag{29}
\]

where \( \delta \) is a constant, typically \( \delta = 1 \).

Furthermore, from (5) it can be shown that in the AOB survival equilibrium point the effluent ratio is

\[
\frac{S_{\text{NO}_2}^f}{S_{\text{NH}_3}^f} = \left(\frac{\mu_1 - D_A}{\mu_2} + \frac{k_{11}}{k_{21}}\right) - D_A k_{\text{NH}_3} \frac{k_{22}}{k_{12}} \tag{30}
\]

Then, by combining (29) and (30), the AOB dilution rate that makes the effluent ratio \( \delta \) can be cleared:

\[
D_A^* = \left(\frac{k_{11} S_{\text{NO}_2}^f + k_{22} S_{\text{NH}_3}^f}{k_{22}}\right) \frac{k_{21}}{k_{11} + k_{21}} = \mu_1 \left(\frac{k_{11} S_{\text{NO}_2}^f + k_{22} S_{\text{NH}_3}^f}{k_{22} + k_{11} S_{\text{NH}_3}^f}\right) \tag{31}
\]
obtained as explained above with (31) and solving (28b) for \( D_A \) (recall that \( S_{NH3}^e \) depends on \( D_A \)). Then, the bounds for dilution rate \( D \) are:

\[
\frac{D_{min}}{\alpha} < D < \frac{D_{max}}{\alpha}.
\]

(34)

The dilution rate obtained in (33) cannot be outside these limits. If that happens, the closest dilution rate between the limits is applied.

The proposed feeding strategy is simulated for process (1) using the parameters in Table 1. The dilution rates are as explained above with \( \alpha = 0.5 \). The desired effluent ratio is \( \delta = 1 \). Multiple simulations were performed, each with different influent ammonium and nitrite concentrations: \( S_{NH3}^f \) takes the values 10, 32, 55, 77.5 and 100 mol m\(^{-3}\) and \( S_{NO2}^f \) takes the values 1, 2, 3, 4 and 5 mol m\(^{-3}\).

The simulation results are shown in Figure 1. The top graph shows the AOB concentrations the middle graph the NOB concentrations and the bottom graph the nitrite to ammonium ratio.

![Fig. 1. Multiple simulations for the proposed control strategy.](image)

Each group of curves corresponds to a different influent ammonium concentration, and each curve in the groups to different nitrite concentrations. Despite influent ammonium concentration is different in each simulations, the objectives are fulfilled in all cases: AOB survives, NOB is repressed (goes to zero) and the effluent ratio goes to the desired value. This also indicates that the dilution rate (33) fulfills the restrictions (34). The major effect of influent ammonium concentration can be noticed on the steady state AOB concentration and the convergence rate of the effluent ratio. In the simulations, for higher influent ammonium concentrations, the equilibrium AOB concentration is higher but convergence rate of the ratio is lower.

The proposed feeding strategy is an open loop type, since only on-line information of the input is used (influent ammonium and nitrite concentrations). For that reason, it is not expected to provide a strong disturbance rejection capacity. Disturbances caused my model uncertainty will cause a miscalculation of the optimal AOB dilution rate, which would cause errors in the dilution rate. However, this problem can be solved by the addition of a feedback loop with the effluent ratio, while keeping the proposed feeding strategy as a feed-forward action. Moreover, if confidence intervals are available for the model parameters, the restrictions on the dilution rate (33) can be calculated using the parameter values which make them the most conservative. In this way, stability guarantees are preserved.

5.1 Simulation results

To illustrate these results, suppose that the stream flow rate can be manipulated and that the bacteria are immobilized in a support where there is a detachment rate \( \alpha \) such that \( D_A = D_N = \alpha D \) where \( \alpha < 1 \) as in Bernard et al. (2001). Then, the dilution rate of the system can be obtained as

\[
D = \frac{D_A}{\alpha}.
\]

(33)

The maximum and minimum bounds for the bacteria dilution rate \( (D_{min}^a \) and \( D_{max}^a) \) are obtained from (28) and solving (28b) for \( D_A \) (recall that \( S_{NH3}^e \) depends on \( D_A \)). Then, the bounds for dilution rate \( D \) are:

\[
\frac{D_{min}}{\alpha} < D < \frac{D_{max}}{\alpha}.
\]

(34)

Or equivalently, which is the influent ammonium concentration that makes the effluent ratio \( \delta \):

\[
S_{NH3}^{f} = \left( \frac{k_{11} \delta + 1}{k_{21}} \right) S_{NH3}^{e} - S_{NO2}^{f} \frac{k_{11}}{k_{21}}
\]

(32)

Then, depending on the available actuators, either (31) or (32) can be used to calculate the input of the process depending on the influent stream composition. This can be used as an open loop control or a feed-forward action.

6. CONCLUSIONS

Restrictions on the process operating parameters were successfully found aimed at making AOB survive while...
represing NOB. The results showed that only one equilibrium point of the process can be feasible and stable for a given set of parameters. Then, only the restrictions from the desired operating point have to be taken into account.

From these results, values for the dilution rate of the process and the influent ammonium can be defined in order to get a given nitrite to ammonium ratio in the effluent. These values along with the restrictions proved to be useful to define an open loop feeding law to repress NOB and achieve the desired effluent. The simulation results successfully showed the validity of the control law in multiple scenarios, making it a good candidate for further development and testing.

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Appendix A. STABILITY OF THE AOB AND NOB SURVIVAL EQUILIBRIUM POINT

The Jacobian matrix at equilibrium $P_e$ is:

$$
\begin{pmatrix}
-D - k_{11}\mu_1'(S_{NH3})^2X_{NOB} & -k_{12}\mu_2'(S_{NO2})^2X_{NOB} & -k_{11}D_A - k_{12}D_N \\
-k_{21}\mu_1'(S_{NH3})^2X_{AOB} & -D - k_{22}\mu_2'(S_{NO2})^2X_{AOB} & -k_{21}D_A - k_{22}D_N \\
\mu_1'(S_{NH3})X_{AOB} & 0 & 0 \\
0 & \mu_2'(S_{NO2})X_{NOB} & 0 \\
0 & 0 & 0
\end{pmatrix}
$$

The eigenvalues of that matrix are difficult to obtain analytically. However, it is enough to show that the eigenvalues are located in the left hemiplane of the complex plane. Assuming feasible equilibrium points, $X_{AOB}^{eq} > 0$ and $X_{NOB}^{eq} > 0$. Then, the trace and determinant of $J$ are:

$$\text{trace}(J) = -2D - k_{11}\mu_1'X_{AOB}^{eq} - k_{22}\mu_2'X_{NOB}^{eq} < 0$$

$$\text{det}(J) = \mu_1'\mu_2'(k_{12}k_{21} + k_{11}k_{22})D_AD_NX_{AOB}^{eq}X_{NOB}^{eq} > 0$$

First, since the trace is the sum of all eigenvalues, that means that there is at least one eigenvalue with negative real part. Secondly, since the determinant is equivalent to the product of all eigenvalues, it can be concluded that there are either two or four eigenvalues with negative real part. Finally, applying the Routh-Hurwitz theorem to the characteristic polynomial of $J$:

$$\Delta s = a_4s^4 + a_3s^3 + a_2s^2 + a_1s + a_0$$

where

$$\begin{align*}
a_4 &= 1 \\
a_3 &= \gamma_1 + 2D \\
a_2 &= \gamma_1D + \gamma_2 + \gamma_3 + D^2 \\
a_1 &= \gamma_2(D_A + D_N) + \gamma_3D \\
a_0 &= \gamma_2D_AD_N
\end{align*}$$

and

$$\begin{align*}
\gamma_1 &= k_{11}\mu_1'X_{AOB}^{eq} + k_{22}\mu_2'X_{NOB}^{eq} > 0 \\
\gamma_2 &= (k_{12}k_{21} + k_{11}k_{22})\mu_1'X_{AOB}^{eq}\mu_2'X_{NOB}^{eq} > 0 \\
\gamma_3 &= k_{11}\mu_1'X_{AOB}^{eq}D_A + k_{22}\mu_2'X_{NOB}^{eq}D_N > 0.
\end{align*}$$

The corresponding Routh-Hurwitz tabulation is:

$$
\begin{array}{c|cccc}
\text{s}^4 & a_4 & a_3 & a_2 & a_1 & a_0 \\
\text{s}^3 & a_4 & a_3 & a_2 & a_1 & a_0 \\
\text{s}^2 & a_4 & a_3 & a_2 & a_1 & a_0 \\
\text{s}^1 & a_4 & a_3 & a_2 & a_1 & a_0 \\
\text{s}^0 & a_4 & a_3 & a_2 & a_1 & a_0
\end{array}
$$

where

$$\begin{align*}
a_1 &= (a_3a_2 - a_4a_1)/a_3 = [\gamma_2D + \gamma_3D + 2D^2 + \gamma_1(\gamma_2 + \gamma_3 + 3D^2)] \\
&+ \gamma_2(2D - D_A - D_N)/[\gamma_1 + 2D] \\
\beta_1 &= (a_1a_4 - a_3a_2)/a_3 = [\gamma_2^2D^2 + \gamma_3^2D^2 + \gamma_1^2D + D_A + D_N] + \gamma_2D(2D + D_A + D_N)) \\
&+ (\gamma_2^2(2DDA + 2DDN - D_A^2 - D_N^2 - 2DA_DN)) \\
&+ \gamma_1^2(2DDA + 2DDN - D_A_DN) + \gamma_1\gamma_2(3DD^2A + 3DD^2N - 4DDA_DN) \\
&+ \gamma_2^2D^2(2DDA + 2DDN - 4DA_DN))/[\gamma_1 + 2D].
\end{align*}$$

It is easy to see that $a_4 > 0$, $a_3 > 0$, $a_0 > 0$. To see that $a_1 > 0$ and $\beta_1 > 0$ note that $D > D_A$ and $D > D_N$. Hence, since there is no sign change in the coefficients in the first column of the Routh tabulation, all the eigenvalues of $J$ evaluated in a feasible $P_x$ have negative real part.