

Kinetics of nitrogen removal in anoxic-anaerobic-aerobic process

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Available online 15 June 2015**Abstract**

Nitrogen removal from wastewater is important to protect the receiving waters from the objectionable aquatic plant growth. Biological nitrogen removal has received considerable interest in recent years. Anoxic-anaerobic-aerobic process was designed and operated to achieve high removals of carbon, nitrogen, and phosphorus from municipal and industrial wastewaters without the addition of chemicals. The research project was to develop the biological kinetic coefficients of nitrogen removal. To determine the coefficients, the mean cell residence time was varied and were operated at five different MLVSS concentrations: 3652, 3315, 2657, 2064, and 1490 mg/L. The theoretical model used in this paper is a Monod-type expression. Nitrification and denitrification kinetic coefficients (Y_N , $k_{d,N}$, K_N , k_N , Y_{DN} , $k_{d,DN}$) were analyzed. Nitrification kinetic coefficients (Y_N , $k_{d,N}$, K_N , and k_N) were 2.28, 0.02, 0.19, and 0.11 respectively. Denitrification kinetic coefficients (Y_{DN} and $k_{d,DN}$) were 1.70 and 0.03 respectively. The relationship between specific denitrification rate and substrate utilization rate for the anoxic stage were also studied.

Keywords: kinetics, anoxic, nitrogen, nitrification, denitrification, kinetic coefficients**บทคัดย่อ**

การกำจัดสารไนโตรเจนออกจากน้ำเสียเป็นสิ่งสำคัญเพื่อป้องกันจากการขยายพืชน้ำในแหล่งน้ำต่าง ๆ วิธีการกำจัดสารไนโตรเจนด้วยวิธีทางชีวภาพเป็นวิธีที่มีความสนใจในการนำมาใช้ในทางปฏิบัติในช่วงปัจจุบัน กระบวนการบำบัดน้ำเสียแบบแอน็อกซิก-แอนแอโรบิก-แอโรบิก ได้ถูกออกแบบและดำเนินการระบบเพื่อให้สามารถกำจัดสารคาร์บอน ไนโตรเจน และฟอสฟอรัสออกจากน้ำเสียทั้งจากชุมชนและโรงงานอุตสาหกรรมโดยไม่ต้องใช้สารเคมีในการช่วยบำบัดน้ำเสีย งานวิจัยนี้ได้พัฒนาค่าสัมประสิทธิ์จลนศาสตร์ชีวภาพของการกำจัดไนโตรเจน โดยได้ทำการปรับเปลี่ยนค่าอายุสลัดจ์ต่าง ๆ และได้ใช้ค่าเอ็มแอลวีเอสเอสต่าง ๆ ทำค่าได้แก่ 3652, 3315, 2657, 2064 และ 1490 มก./ล ในบทความนี้ได้พัฒนาค่าสัมประสิทธิ์จลนศาสตร์ชีวภาพของการกำจัดไนโตรเจน งานวิจัยนี้ได้ใช้สมการโมโนต์ในการสร้างแบบจำลองทางทฤษฎีขึ้นมาเพื่อใช้ในการวิเคราะห์ ค่าสัมประสิทธิ์จลนศาสตร์ของไนตริฟิเคชันและดีไนตริฟิเคชัน (Y_N , $k_{d,N}$, K_N , k_N , Y_{DN} , $k_{d,DN}$) ได้ถูกวิเคราะห์หาขึ้นมาจากแบบจำลองทางทฤษฎีที่ถูกสร้างขึ้น ค่าสัมประสิทธิ์จลนศาสตร์ของไนตริฟิเคชัน (Y_N , $k_{d,N}$, K_N , k_N) ที่ได้เท่ากับ 2.28, 0.02, 0.19 และ 0.11 ตามลำดับ ค่าสัมประสิทธิ์จลนศาสตร์ของดีไนตริฟิเคชัน (Y_{DN} , $k_{d,DN}$) ที่ได้เท่ากับ 1.70 และ 0.03 ตามลำดับ นอกจากนี้ได้ศึกษาหาความสัมพันธ์ระหว่างค่าอัตราการเกิดดีไนตริฟิเคชันจำเพาะและค่าอัตราการใช้สับสเตรตสำหรับช่วงแอน็อกซิกด้วย

คำสำคัญ: จลนศาสตร์, แอน็อกซิก, ไนโตรเจน, ไนตริฟิเคชัน, ดีไนตริฟิเคชัน, ค่าสัมประสิทธิ์จลนศาสตร์**1. Introduction**

Increasingly stringent regulations on nitrogen and phosphorus removal from wastewater have spurred global efforts to develop and implement cost-effective engineered biological nutrient removal (BNR) technologies at wastewater treatment plants (Ahn et al., 2010, p. 4505). The biological removal of nitrogen is widely used for treatment of domestic as well as complex industrial wastewaters. A number of techniques have been proposed in recent years aimed at modification of the conventional activated sludge process to improve removal of nitrogen and/or phosphorus (Qasim,

1999, p. 430). These modifications have attempted to provide more economical and higher removal of nitrogen and/or phosphorus by optimizing the various factors controlling the microbiological process. Combined nitrification-denitrification can be achieved in a series of reactors that create aerobic and anoxic conditions. Raw wastewater is utilized as an external organic carbon source. Under aerobic or anoxic conditions, ordinary heterotrophic organisms use organic substrates as energy and carbon sources. The yield of biomass growth is the fraction of substrate that is used as a carbon source to produce biomass (Hauduc et al., 2013, p. 24). The anoxic-

anaerobic-aerobic process used in this study was developed by the University of Texas at Arlington. Over the past 80 years, a number of processes have been used for the removal of nitrogen from wastewater. Such as the classical biological nitrification-denitrification process for nitrogen removal which was first applied in wastewater in 1940 (Sutton, Murphy, & Dawson, 1975, p. 123). All of the biological nitrogen removal processes include an aerobic zone in which biological nitrification converts $\text{NH}_4\text{-N}$ to $\text{NO}_2\text{-N}$ and $\text{NO}_3\text{-N}$. Anoxic time must also be included to provide biological denitrification to complete the objective of total nitrogen removal by both $\text{NH}_4\text{-N}$ oxidation and $\text{NO}_3\text{-N}$ and $\text{NO}_2\text{-N}$ reduction to nitrogen gas (Metcalf & Eddy Inc. & AECOM, 2014, p. 797). Chemical oxygen demand (COD) removal takes place in all of the different zones, while the nitrification process is carried out in only the aerobic zone. Nitrogen removal proceeds by the denitrification of the generated nitrite and nitrate in the microaerophilic and anoxic zones (Alimahmoodi, Yerushalmi, & Mulligan, 2012, p. 1136). Nitrifying organisms are present in almost all aerobic biological treatment processes. The ability of activated sludge to nitrify was correlated to the BOD_5/TKN ratio (Metcalf & Eddy Inc., 2003, p. 758). The influent wastewater characteristics are important in affecting biological denitrification rates in the anoxic system. Nitrate reduction is dependent on having sufficient electron donors, so there must be a sufficient amount of influent biochemical oxygen demand (BOD) relative to the amount of nitrogen to be removed. As a rule of thumb, an influent BOD to TKN ratio of 4/1 is necessary to provide a sufficient amount of electron donor (Randall, Barnard, & Stensel, 1992, p. 55). The denitrification rate generally follows a zero order reaction when carbon is not a limiting factor (Beccari, Passino, Ramadori, & Tandoi, 1983, p. 58). The rate of denitrification in the anoxic zone is affected by the BOD concentration in the influent wastewater and the MLVSS concentration (Metcalf & Eddy Inc. & AECOM, 2014, p. 797). The higher concentration of active mass of the mixed liquor at the short solids retention time (SRT) resulted in good denitrification in the unaerated sections of the channels (Barnard, 1998, p.330). Values of the denitrification rate have been used to characterize denitrification rates in different anoxic systems as well as to evaluate the effect of different external carbon sources. In a single-sludge system with an excess organic carbon source, the denitrification rate

ranges from 0.075-0.115 g $\text{NO}_3\text{-N/g}$ MLVSS.d (Qasim, 1999, p. 434).

2. Objectives

The objective of this study was to develop the biological kinetic coefficients for removal of nitrogen in anoxic-anaerobic-aerobic process. Nitrification kinetics and denitrification kinetics were analyzed in this process. Mathematical models of the process were developed and included BOD removal, nitrification and denitrification. In this study three bench-scaled completely mixed continuous flow reactors were used. The developed kinetic coefficients were available for designing the treatment process effectively. The experimental program dealt with biological nutrient removal studies without chemical addition and developed a treatment process that offered high nitrogen and phosphorus removal. As part of the bench scale reactor study, the program also observed the concentrations of nitrogen and phosphorus throughout the treatment process at various MLSS concentrations. The research was focused on single-sludge suspended growth system that combined the processes of carbon oxidation, nitrification, denitrification, and release-uptake of phosphorus in the three reactors system. The final goal of this research work was to determine the relationship between specific denitrification rate and substrate utilization rate for the anoxic stage.

3. Materials and methods

3.1 Experimental set-up

The biological nutrient removal system utilizes a series of three reactors to accomplish the desired removals. The wastewater and return sludge are mixed in the first reactor in which an anoxic condition is maintained. The contents of the first reactor are continuously discharged into a second reactor which maintains an anaerobic environment. The contents from the second reactor are then fed into the third reactor which maintains an aerobic condition. The schematic flow diagram is given in Figure 1. Three plexiglass completely mixed continuous-flow reactors for biological nutrient removal study were used in the experimental program. The operating features of the reactors are summarized in Table 1. The mean cell residence time was used as a variable to achieve different operating conditions. This was done by operating the process at five different effective MLVSS concentrations: 3652, 3315, 2657, 2064, and 1490

mg/L. Each MLVSS concentration was maintained for several weeks to obtain the stable values of substrate removal, and sludge growth rates. Excess MLVSS from the system was wasted at the same time daily. The amount wasted was based on the effective MLVSS analysis (see equation (1)) of the entire system and the total suspended solids lost in the effluent for that day. The amount of biomass

produced each day was equal to the sum of the biomass intentionally wasted and the biomass lost in the effluent. As mentioned earlier the system was operated at five effective MLVSS concentrations. All other parameters such as influent flow and characteristics BOD, total nitrogen, total phosphorus, recycle rate, and air flow rate were kept constant for all values of effective MLVSS concentrations.

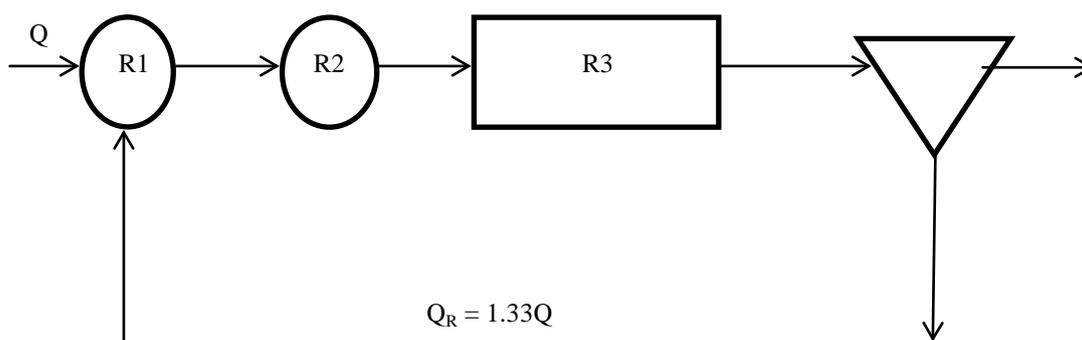


Figure 1 Anoxic-Anaerobic-Aerobic process (R1=Anoxic reactor, R2=Anaerobic reactor, R3=Aerobic reactor)

Table 1 Operating features of the reactors

| Conditions | Values |
|---------------------------------|------------|
| Substrate feed rate | 0.03 L/min |
| Recirculation ratio (Q_R/Q) | 1.33 |
| Operating temperature | 20-25°C |
| Anoxic reactor: | |
| Liquid volume | 3.19 L |
| Detention time w/o return flow | 1.77 h |
| Detention time with return flow | 0.76 h |
| Anaerobic reactor: | |
| Liquid volume | 1.64 L |
| Detention time w/o return flow | 0.91 h |
| Detention time with return flow | 0.39 h |
| Aerobic reactor: | |
| Liquid volume | 10.38 L |
| Detention time w/o return flow | 5.77 h |
| Detention time with return flow | 2.47 h |
| DO concentration | 5-7 mg/L |

$$\text{Effective MLVSS} = \frac{C_1 V_1 + C_2 V_2 + C_3 V_3}{V_1 + V_2 + V_3} \quad (1)$$

$C_1, C_2,$ and C_3 = Measured MLVSS concentrations in each reactor, mg/L
 $V_1, V_2,$ and V_3 = Volume of each reactor, L

4. Results

4.1 Characteristics of influent feed

The wastewater feed was obtained from a municipal sewer. This wastewater contained lower concentrations of BOD, phosphorus, and nitrogen than those in medium strength wastewater. Therefore small amounts of powdered milk, ammonium chloride, and dipotassium hydrogen phosphate were added to enrich the wastewater. The

quality of enriched domestic wastewater for use in this research program is summarized in Table 2.

Nitrification and denitrification are important microbiological reactions of nitrogen. In this work, the kinetics of these reactions has been investigated based on a Monod-type expression. The kinetic constants and yield coefficients were evaluated for both of these reactions. The experimental results of the system operation are given in Table 3.

Table 2 Quality of enriched domestic wastewater

| Parameters | Average enriched domestic wastewater |
|--------------------------------|--------------------------------------|
| BOD ₅ , mg/L | 344 |
| COD, mg/L | 543 |
| NH ₃ -N, mg /L as N | 30 |
| TKN, mg /L as N | 49 |
| ORG-N, mg /L as N | 19 |
| ORTHO-P, mg/L as P | 8.3 |
| TP, mg/L as P | 12.3 |
| pH | 7.8 |

Table 3 Experimental results of the system operation

| Parameters | MLVSS maintained in system, mg/L | | | | |
|---|----------------------------------|-------|-------|-------|-------|
| | 3652 | 3315 | 2657 | 2064 | 1490 |
| θ_c , d | 11.70 | 13.80 | 16.20 | 15.30 | 5.50 |
| BOD _{in} ¹ , mg/L | 459 | 427 | 348 | 320 | 320 |
| BOD _{ef} ² , mg/L | 5 | 4 | 3 | 5 | 5 |
| NH ₄ -N _{in} ¹ , mg/L as N | 30 | 31 | 28 | 26 | 30 |
| NH ₄ -N _{ef} ² , mg/L as N | 0.1 | 0.1 | 0.1 | 0.2 | 0.4 |
| NO _{3aerobic} , mg/L as N | 14.58 | 11.99 | 15.36 | 11.50 | 10.90 |
| NO _{3anoxic} , mg/L as N | 0.22 | 0.45 | 0.26 | 0.49 | 0.15 |

¹_{in} means soluble influent, ²_{ef} means soluble effluent

4.2 Determination of nitrification kinetics

The form of the rate expressions for nitrification growth rates, based on saturation kinetics for ammonia oxidation as given in equation (2) to (5), assuming excess DO is available (Metcalf & Eddy Inc., 2003, p. 759). Using activated sludge mixed liquor, Wild's 1971 study (as cited in Randall et al., 1992, p. 34) found that nitrification rates were not affected by DO concentrations above 1.0 mg/L. In this research project, the aerobic reactor was operated with DO concentrations between 5-7 mg/L.

$$\frac{1}{\theta_c} = Y_N U_N - k_{d,N} \quad (2)$$

θ_c = Sludge age, d

Y_N = Maximum cell yield coefficient for nitrifiers, kg VSS/kg NH₄-N

U_N = Specific substrate utilization rate for nitrifiers, per day

$k_{d,N}$ = Endogenous decay coefficient for nitrifiers, per day

$$\frac{1}{U_N} = \frac{K_N}{k_N N_{ef}} + \frac{1}{k_N} \quad (3)$$

K_N = Half- saturation coefficient for ammonia nitrogen limited reaction, mg as N/L

k_N = Maximum rate of ammonia nitrogen utilization per unit mass of VSS, per day

N_{ef} = Ammonia nitrogen in effluent, mg as N/L

$$k_N = \frac{\mu_{max,N}}{Y_N} \quad (4)$$

$\mu_{max,N}$ = Maximum specific growth rate for nitrifiers, per day

$$\theta_{c,N}^{min} = \frac{1}{\mu_{max,N} - k_{d,N}} \quad (5)$$

$\theta_{c,N}^{min}$ = Minimum SRT for nitrifiers, day

Y_N and $k_{d,N}$ were determined by linear regression fits of the data points in Table 4 using equation (2). K_N and k_N were determined by linear regression fits of the data points in Table 4 using equation (3). These nitrification kinetic coefficients for the anoxic-anaerobic-aerobic process are summarized in Table 5.

The half-saturation coefficient (K_N) is an important kinetic parameter in the application of the Monod model to nitrification reactor design. The half-saturation coefficient found for nitrification is generally very low compared to values commonly reported for heterotrophic organisms. Based on full scale plant performance, K_N values between 0.30-0.70 mg as N/L may be expected (Metcalf & Eddy Inc. & AECOM, 2014, p. 754). It appears that maximum specific growth rates for nitrifiers ($\mu_{max,N}$) at 10-20°C are in the range of 0.1-0.7 per day (Randall et al., 1992, p. 31). $\mu_{max,N}$ and $\theta_{c,N}^{min}$ are calculated from Equation (4) and (5) which are 0.25 per day and 4.35 day respectively.

Table 4 Calculated values for determination of nitrification kinetic coefficients

| Parameters | MLVSS maintained in system, mg/L | | | | |
|-----------------------------|----------------------------------|-------|-------|-------|-------|
| | 3652 | 3315 | 2657 | 2064 | 1490 |
| $1/\theta_c$, 1/d | 0.085 | 0.072 | 0.062 | 0.065 | 0.182 |
| ΔNH_4-N , mg/L as N | 29.9 | 30.9 | 27.9 | 25.8 | 29.6 |
| U_N , 1/d | 0.034 | 0.039 | 0.044 | 0.052 | 0.083 |
| $1/U_N$, d | 29.41 | 25.64 | 22.73 | 19.23 | 12.05 |
| $1/N_{ef}^1$ | 10 | 10 | 10 | 5 | 2.5 |

¹ NH_4-N effluent

Table 5 Nitrification kinetic coefficients for the anoxic-anaerobic-aerobic process

| Nitrification kinetic coefficient parameter | Unit | Value |
|--|--------------------|-------------------|
| Maximum cell yield coefficient for nitrifiers, Y_N | kg VSS/kg NH_4-N | 2.28 ¹ |
| Endogenous decay coefficient for nitrifiers, $k_{d,N}$ | per day | 0.02 ¹ |
| Half-saturation coefficient for ammonia nitrogen limited reaction, K_N | mg as N/L | 0.19 ² |
| Maximum rate of ammonia nitrogen utilization per unit mass of VSS, k_N | per day | 0.11 ² |

¹Linear correlation coefficient (R)= 0.88 , ²R= 0.92

4.3 Determination of denitrification kinetics

For biological denitrification, based on saturation kinetics for an anoxic suspended growth process, the biokinetic equation is given in equation (6). A relationship between sludge age and specific denitrification rate can then be used to determine the maximum cell yield coefficient for denitrification and the endogenous decay coefficient for denitrification.

$$\frac{1}{\theta_c} = Y_{DN} U_{DN} - k_{d,DN} \quad (6)$$

θ_c = Sludge age, d

Y_{DN} = Maximum cell yield coefficient for denitrifiers, kg VSS/kg NO_3-N

U_{DN} = Specific denitrification rate, g NO_3-N/g

MLVSS.d

$k_{d,DN}$ = Endogenous decay coefficient for denitrifiers, per day

Specific denitrification rate is the nitrate reduction rate in the anoxic tank normalized to the MLVSS concentration. Nitrate serves as an electron acceptor in the same way as oxygen from a biokinetics perspective, and thus the denitrification rate is proportional to the substrate utilization rate. The specific denitrification rate is determined by equation (7). The specific denitrification rates (U_{DN}) in the range of 0.047-0.097 g NO_3-N/g MLVSS.d are shown in Table 6. These values indicated specific denitrification rates that ranged from 0.4 g NO_3-N/g MLVSS.d for the first stage of a high-loaded four-

stage pre-anoxic zone, to 0.05 g NO₃-N/g MLVSS.d for a single-stage anoxic zone, to the COD loading (Panzer, Komanowsky, & Senske, 1980, p. 108).

$$U_{DN} = \frac{\Delta NO_3}{XV_{anoxic}} \quad (7)$$

ΔNO_3 = Nitrate removed, g N/d
 X = MLVSS in the system, mg/L
 V_{anoxic} = Anoxic tank volume, m³

The specific denitrification rate (U_{DN}) can be expected to be proportional to the food of the MLVSS ratio for the anoxic stage (see equation(8)). The value for F/M_{anoxic} is based on the influent BOD and controlled MLVSS concentration. Once these are known, the value for U_{DN} can be calculated.

$$U_{DN} = 0.00156 + 0.033(F/M_{anoxic}) \quad (8)$$

F/M_{anoxic} = g BOD applied/g MLVSS.d in the anoxic tank

R of Equation (8) is 0.84. Equation (8) is based on data collected for anoxic/anaerobic/aerobic processes at room temperatures in the range of 20 - 25°C. The empirical equation provides only a rough estimate of the specific denitrification rate. Based on observed denitrification rates in pilot plant and full scale plants, the empirical relationship can be described by equation (9) (Burdick, Refling, & Stensel, 1982, p. 1082).

$$U_{DN} = 0.029 + 0.03(F/M_{anoxic}) \quad (9)$$

Table 6 Calculated values for determination of denitrification kinetic coefficients

| Parameters | MLVSS maintained in system, mg/L | | | | |
|---------------------------|----------------------------------|-------|-------|-------|-------|
| | 3652 | 3315 | 2657 | 2064 | 1490 |
| MLVSS, g | 11.65 | 10.57 | 8.48 | 6.58 | 4.75 |
| ΔNO_3 , mg/L as N | 14.36 | 11.54 | 15.10 | 11.01 | 10.75 |
| ΔNO_3 , g N/d | 0.62 | 0.50 | 0.65 | 0.48 | 0.46 |
| $1/\theta_c$, 1/d | 0.085 | 0.072 | 0.062 | 0.065 | 0.182 |
| U_{DN} , 1/d | 0.053 | 0.047 | 0.077 | 0.073 | 0.097 |
| BOD applied, g/d | 19.83 | 18.45 | 15.03 | 13.82 | 13.82 |
| F/M_{anoxic} | 1.70 | 1.75 | 1.77 | 2.10 | 2.91 |

Table 7 Denitrification kinetic coefficients for the anoxic-anaerobic-aerobic process

| Denitrification kinetic coefficient parameters | Unit | Value |
|---|------------------------------|-------------------|
| Maximum cell yield coefficient for denitrifiers, Y_{DN} | kg VSS/kg NO ₃ -N | 1.70 ¹ |
| Endogenous decay coefficient for denitrifiers, $k_{d,DN}$ | per day | 0.03 ¹ |

¹ R = 0.67

Y_{DN} and $k_{d,DN}$ were determined by linear regression fits of the data points in Table 6 using equation (6). These denitrification kinetic coefficients for the anoxic-anaerobic-aerobic process are given in Table 7.

Nitrified influent is fed to a mixed anoxic tank along with the wastewater, which is an external carbon source. Denitrification kinetic coefficients for the anoxic-anaerobic-aerobic process, based on BOD in the wastewater without methanol addition, are given in Table 7. Endogenous respiration creates a demand for nitrate in addition to that caused by substrate utilization and oxidation. This reaction occurs in the mixed liquor of the anoxic tank and is

at a much lower rate than the denitrification rate caused by substrate utilization. It appears that endogenous decay coefficient for denitrifiers ($k_{d,DN}$), with methanol as the growth substrate at 10-20°C, are in the range of 0.04-0.05 per day (Metcalf & Eddy Inc., 2003, p. 785).

5. Conclusion

The biological kinetic coefficients developed for nitrogen are useful in designing the biological anoxic-anaerobic-aerobic process. Nitrification and denitrification kinetics were developed in this project. Important observations from the research are as follows.

- a) The bench-scale completely mixed continuous flow reactors have to be carried out to ascertain the method's adaptability in practice.
- b) A high degree of denitrification can be achieved in a single-sludge system without the addition of a chemical as an external carbon source. The wastewater serves as the carbon source.
- c) Further investigation is needed whereby return flow may be varied to achieve maximum overall nitrogen removal without decreasing BOD and phosphorus removals.
- d) The reactor sequence is such that the anoxic and anaerobic conditions can be created inside a long conduit or force main if sludge is returned into the pipe. This part of the treatment therefore can be achieved in a pipeline.
- e) For the five different effective MLVSS concentrations studied in this work (3692, 3315, 2657, 2064 and 1492 mg/L), BOD and nitrogen removals were not significantly different.
- f) The specific denitrification rate (U_{DN}) is proportional to the food to MLVSS ratio (F/M_{anoxic}) for the anoxic stage.
- g) The analyzed kinetic coefficients are limited for our biological nutrient removal process.

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