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Energy efficient treatment of A-stage effluent: pilot-scale experiences with shortcut nitrogen removal

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Abstract

Energy autarky of sewage treatment plants, while reaching COD and N discharge limits, can be achieved by means of shortcut N-removal. This study presents the results of a shortcut N-removal-pilot, located at the biological two stage (high/low rate) wastewater treatment plant of Breda, NL. The pilot treated real effluent of a high rate activated sludge (COD/N=3), fed in a continuous mode at realistic loading rates (90-100 g N/m^3/d). The operational strategy, which included increased stress on the sludge settling velocity, showed development of a semi-granular sludge, with average particle size of 280 µm (Ø$_{4,3}$), resulting in increased suppression of nitrite oxidizing bacteria. The process was able to remove part of the nitrogen (51±23%) over nitrite, with COD/N removal ratios of 3.2±0.9. The latter are lower than the current operation of the full-scale B-stage in Breda (6.8-9.4), showing promising results for carbon efficient N-removal, while producing a well settling sludge (SVI$_{30}$<100 mL/g).

Keywords: Energy-positive wastewater treatment, granulation, mainstream nitrogen removal, nitritation/denitritation, partial nitritation/anammox, shortcut nitrogen removal

INTRODUCTION

Worldwide, increasing energy prices and more stringent effluent norms push the boundaries of the current state-of-the-art sewage treatment plants. Energy self-sufficiency and autarky of sewage treatment plants is possible by means of more efficient usage of carbon and nutrients present in wastewater. To achieve this, a two stage process can be implemented to valorize wastewater carbon constituents. In a first step (A-stage), the harvesting of COD is maximized to produce chemicals or biogas, and in a second step (B-stage), nitrogen is removed from the remaining carbon-poor wastewater-stream. In order to optimize carbon capture, the COD/N ratio is often too low to treat through conventional nitrification/denitrification (N/DN). More carbon-efficient removal of nitrogen can be achieved by usage of a shortcut route based on nitritation/denitritation (Nit/DNit) and/or partial nitritation/anammox (PN/A). Out of these two, the most desired process is the one that reaches discharge limits for COD and N at minimal energy expenditure (Verstraete & Vlaeminck 2011).

PN/A, a technology that removes nitrogen without the need of organic carbon, is a state-of-the-art technology for N-rich wastewaters at higher temperatures (>500 mg NH_4^+-N/L, > 25 °C), with over 100 full-scale installations being built by 2014 (Lackner *et al.* 2014)**.** Domestic sewage however has typical lower nitrogen concentrations (30-40 mg N/L) and fluctuating temperatures due to seasonal variations, this in a range of 8-25 °C in moderate climate regions like Western Europe. Furthermore, the robustness of the technology will be challenged due to events of rainfall, thaw, and failing of pretreatment steps like the A-stage.

Recent studies show promising results for the utilization of PN/A under moderate climate conditions in the laboratory (De Clippeleir *et al.* 2013; Gilbert *et al.* 2014b; Lotti *et al.* 2014b). Yet, suppression of nitrite oxidizing bacteria (NOB) in these systems remains challenging. Typical suppression strategies of NOB include the use of inhibitory levels of free ammonia (FA) in the reactor (Anthonisen *et al.* 1976), however, due to lower N concentrations of domestic sewage, this strategy cannot be used. Therefore NOB should be suppressed kinetically (e.g. by limiting their access to substrate).

Kinetic NOB suppression can be realized by **1)** utilizing intermittent aeration while inducing a nitratational lag (Kornaros *et al.* 2010; Gilbert *et al.* 2014a), **2)** stimulating aerobic ammonium oxidizing bacteria (AerAOB) by residual levels of NH_4^+ (Regmi et al. 2014), while **3)** avoiding over-aeration (= avoiding the availability of oxygen for NOB when no ammonium is present) by aeration duration control (Blackburne *et al.* 2008a), **4)** applying higher (resp. lower) dissolved oxygen (DO) levels to outcompete *Nitrospira* spp. (resp. *Nitrobacter* spp.) vs. AerAOB (Sliekers *et al.* 2005; Blackburne *et al.* 2008b) or **5)** increasing the competition for nitrite at lower DO levels with anoxic pathways in larger aggregates (anammox, denitritation) (Vlaeminck *et al.* 2010). Another strategy uses **6)** free nitrous acid (FN/A), produced in a sidestream contact tank to inhibit NOB (Wang *et al.* 2014).

To realize a one stage shortcut nitrogen removal reactor under these conditions, a separate sludge retention time (SRT) for the different pathways is warranted, as an add-on for the kinetic NOB suppression: a high SRT for aerobic and anoxic ammonium oxidation and a low SRT for nitrite oxidation and aerobic heterotrophs. This can be realized by **a)** induction of granulation or addition of a carrier material to retain bacteria growing in biofilms, **b)** utilization of sieves or tilted plate separators for advanced sludge selection and **c)** application of shear to wash-off unwanted bacterial groups from the aggregates by for example hydrocyclones (Wett *et al.* 2010). In this way, slower growing organisms like the anoxic ammonium oxidizing bacteria (AnAOB), typically growing in biofilms, can increase their SRT in the system, while retaining a short enough SRT to remove NOB and heterotrophs out of the system.

The study presents the first results of the UNAS (Upflow New Activated Sludge) pilot, located at the A/B wastewater treatment plant of Breda, NL. In this WWTP, the A stage is a high-rate activated sludge reactor, with an anoxic zone to denitrify returned nitrate from the B stage, followed by an aerobic zone to redirect as much as possible the organic carbon to the digester. The B stage is a conventional, i.e. low-rate, nitrification/denitrification stage.

In the pilot, kinetic NOB suppression strategies (**1, 2** and **5**) are combined with a sludge selection strategy based on granulation (**a**) and sieves (**b**) to select for more granular opposite to floccular sludge. The pilot treated real effluent of a high rate activated sludge A-stage (COD/N=3), fed in a continuous mode at realistic loading rates (90-100 g N/m^3/d). The aim of the pilot plant is to achieve a nitrogen shortcut, combined with granulation to efficiently remove nitrogen and carbon from the effluent of the A-stage.

MATERIALS AND METHODS

Reactor

A 7 m^3 (H = 4 m, Ø = 1.495 m) pilot scale reactor was operated at 20°C at the wastewater treatment plant Nieuwveer in Breda, NL. The reactor was inoculated with floccular sludge from a sidestream PN/A treatment plant. The cylindrical reactor consisted of two vertical recycle streams, controlling the upflow velocity separately in the bottom (height 0-3 m) and top (>3m) compartment. Unless mentioned otherwise, velocities during operation were 2.3 m/h for the lower and 0.45 m/h for the upper compartment of the reactor. The effluent was discharged batchwise and pumped over a sieve (Table 1). The retained sludge was transferred to a sludge buffer and transferred back to the reactor in a batchwise manner. The technology was a further development of the New Activated Sludge (NAS®) concept as it was developed by Colsen bv together with Ghent University in 2004 (Desloover *et al.* 2011). The operation of the reactor is depicted in Figure 1.

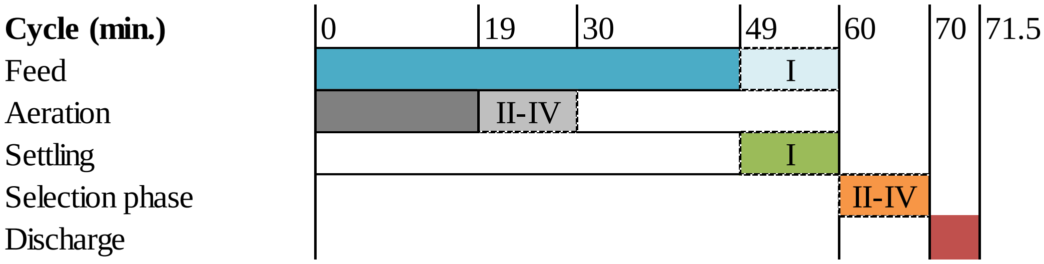


Figure 1. Operational cycle of the pilot. Dashed lines indicate different timings experimented with along the operation of the reactor. The different experimental periods I-IV (see Table 1) are also highlighted.

Influent

The feed of the reactor consisted of the effluent of the A-stage, supplemented with extra ammonium as of ammonium sulfate (days 0-37) or urea (days 38-190). This was done to mimic expected COD/N concentrations of 3 in the influent. Final concentrations can be found in Table 1. The pH was controlled from day 38 onward through addition of Na_2CO_3 to compensate for the extra added N. It was controlled towards the expected pH of 7-7.3, which is the pH of the effluent of the B-stage of Breda, and has the same alkalinity-need as a PN/A process.

Measurements and control

Levels of dissolved oxygen (DO) (S::SCAN oxi::lyser), pH and NH_4^+ (S::SCAN amo::lyser) NO_2^-, NO_3^- (S::SCAN spectro::lyser) were measured on-line in the reactor and DO concentration was controlled by means of a PID controlled setpoint during the aerobic phase in the reactor.

Determination of nitrification kinetics

Procedure

In a mixed 2L reactor, the DO concentration was kept constant by on-off control for at least half an hour (depending on expected nitrogen removal rates). At the start of this timeframe, NH_4^+ (NH_4HCO_3) and NO_2^- (KNO_2) spikes were added to obtain concentrations of resp. 10 mg N/L and 5 mg N/L. Six samples were taken for NH_4^+, NO_2^- and NO_3^- at time intervals between 5 and 12 minutes (again depending on the expected N-removal rate), filtered immediately over 0.45µm filters and stored at 4°C. The rate of decrease/increase in NH_4^+/NO_3^- at a certain DO concentration resulted in a measure for AerAOB and NOB activity respectively.

This procedure was repeated for a total of 6 dissolved oxygen concentrations (2.5, 1.5, 1.0, 0.7, 0.5 and 0.2 mg O_2/L), theoretically all laying on the Monod curve for oxygen. Temperature was kept constant during the whole experiment at 21°C. The pH was controlled at pH=7.5, with solutions of 0.5 M HCl and NaOH respectively. Data collection and control of the DO concentration was done using LabVIEW (National Instruments, USA).

Parameter estimation

To estimate both the oxygen affinity constant (K_s-O_2) and maximum removal rate (R-max) values, the experiment was modeled in Excel by constructing relevant mass balances for the nitrogen species (NH_4^+, NO_2^- and NO_3^-), including conversion of nitrogen species by both AerAOB and NOB. The obtained removal rates at different dissolved oxygen levels were fit to the relevant Monod-equation using minimization of Sum of Squared Errors (SSE), yielding apparent K_s-O_2 an R-max-values. During testing, it was ensured that both NH_4^+, and NO_2^- concentrations remained above 2 times the half saturation coefficient levels, diminishing the influence of the nitrogen Monod-terms on the parameter estimation.

Analyses

For the samples from the pilot installation, NH_4^+, NO_2^-, NO_3^-, total nitrogen (TN), soluble and total chemical oxygen demand (COD) were analyzed on 24h samples with dr. Hach Lange test kits. Total/volatile suspended solids (TSS/VSS) and the sludge volume index after 30 minutes (SVI_{30}) were measured according to the standard methods for examination of water and wastewater (APHA 2003)**.** For the samples from the nitrification kinetics determination, analyses were done using the Nessler method for NH_4^+ (APHA 2003) and using Ion Chromatography for NO_2^- and NO_3^- with a 761 Compact IC, Metrohm, Switzerland.

Particle size distribution was measured using laser diffraction with a Mastersizer S long bench (Malvern, UK), lens 1000F to measure particle sizes in a range of 4-3.5 mm, with 10 000 sweeps and particle obscuration between 10 and 30%. Results were fit to an optical model, code 3PHD and resulted in an average volume weighed particle size $\emptyset_{4,3}$, 10% smallest particles $d_{0.1}$ and 10% largest particles $d_{0.9}$.

Calculations

To assess how much nitrogen is removed over nitrite, the COD/N removal ratio can be compared to the theoretical COD/N need of 4.07 for denitrification (DN) and 2.4 for DNit, including cell growth for typical organic matter, e.g. C_5H_9NO (Mateju *et al.* 1992). With this in mind, following assumptions were made to calculate how much nitrogen was removed over nitrite; **1)** since no nitrite accumulation was measured during the aerobic phase, it can be assumed that all nitrogen removal over nitrite occurred in the aerobic phase **2)** equal COD conversion rates occurred under aerobic as well as under anoxic conditions, **3)** No COD was removed aerobically in the aerobic phase (=worst case scenario) and **4)** from the weighted average of the COD/N removal ratio, the % of nitritation was calculated.

RESULTS AND DISCUSSION

The reported timeframe can be divided into four periods, each with different selection mechanisms to form a better settling sludge with a higher average (volume weighed) particle size. The effects on the sludge characteristics and performance are depicted in Figure 1 and Table 1. In period I, larger particles were selected by increasing the minimum settling velocity (MSV) in m/h over time every cycle (24 times per 24 cycles, x m/h). In period II-IV an extra selection phase was included with a certain frequency (+y times per 24 cycles) and applied settling rate (z m/h).

Table 1. Pilot reactor operational parameters. Minimum settling velocity = Minimum settling velocity (x m/h) applied to the reactor in every cycle (24/24); Selection phase = extra settling rate applied (z m/h), with a frequency of only y/24 cycles; [1] $(NH_4)_2SO_4$ added, [2] Organic nitrogen (Norg) = total (TN) - inorganic nitrogen, [3] mainly derived from urea.

Period		**I**	**II**	**III**	**IV**
Minimum settling velocity (x m/h) **Selection phase (y/24, z m/h)**		**24/24 0.5 --> 0. 9 m/h**	**24/24 0.9-1 m/h** **+1/24 2.5-3 m/h**	**24/24 0.8-1 m/h** **+3/24 3 m/h**	**24/24 0.8 m/h** **+4/24 3 m/h**
Days		0-103	104-122	123-151	152-190
Sieve size (mm)		0.1 (Day 0-70) 0.2 (Day 71-103)	0.2	0.413	0.413
Influent	**TN (mg N/L)**	41.9±5.2	37.6±4.6	40.5±11.2	34.2±4.7
	NH_4^+ (mg N/L)	33.8 1.6 (Day 0-38)[1] 15.9±2.7 (Day 0-103)	14.5±2.6	16.7±6.8	10.8±2.8
	NO_2^- (mg N/L)	0.2±0.1	0.3±0.2	0.2±0.1	0.2±0.1
	NO_3^- (mg N/L)	1.4±0.5	1.1±0.4	0.9±0.2	2.±3.1
	Norg (mg N/L)[2]	5.3±2.3 (Day 0-38) 25.3±3.7 (Day 39-103)[3]	22±4.5[3]	20.82±3.5[3]	20.5±5.5[3]
	COD/TN	3.3±0.9	3.05±0.6	3.2±0.9	2.1±1.5
Reactor	**DO (mg O_2/L)**	0.6-0.8 (Day 0-5) 0.4-0.6 (Day 6-13) 0.3-0.4 (Day 14-103)	0.3-0.4	0.3-0.4 (Day 123-148) 0.6 (Day 149-151)	0.6 (Day 152-158) 0.45 (Day 159-190)
	pH (-)	6.5±0.1 (Day 0-38) 6.8±0.1 (Day 39-103)[1]	6.9±0.2	7.1±0.1	7.3±0.1
	Aerobic fraction (%)	50	45	45	39
	Bv (mg N/m^3/d)	79.6±26.0	107.9±12.7	91.8±20.4	91.9±11.9
	Bx (mg N/gVSS/d)	41.4±15.9	48.0±13	38.3±4.7	39.6±6.1
	Rv/Bv (%)	28±11% --> 51±14	49±14% -52±16	41±2 % --> 34±6	35±15% --> 23±16
	Nitrogen removal over nitrite (%)	62±22.2 (Day 46-103)	51.3±23.1	32.9±15.1	49.3±24

Period I

Selection strategy and overall performance

A first selection strategy was applied by decreasing the settling time from 11 to 0 minutes corresponding to an applied MSV of 0.55 m/h to 0.9 m/h. This resulted in a desirable increase in particle size and decrease in SVI_{30}. Furthermore, the nitrogen removal increased from 27 to 50%, with a decrease in nitrate effluent levels from 25 to 15 mg N/L.

At the start of the experiment, the COD/N removal ratio ranged between 4 and 12, indicating mainly nitrogen removal over nitrate. Slight nitrate accumulation in the aerobic phase showed that NOB were not completely suppressed. However, at a higher selection pressure, the COD/N removal ratio was below 4, clearly indicating the presence of a nitrite shortcut route. From previous assumptions (see material and methods), it could be estimated that 61±22% of the nitrogen removal occurred directly over nitrite.

Evolution of the nitrification kinetics

The evolution of the nitrification kinetic parameters R-max and K_s-O_2 over period I are given in Figure 3. The oxygen affinity indices show that NOB still had an advantage at lower DO concentrations compared to AerAOB, provided there is no limitation in nitrite/ammonium. However, nitrite was only present at low concentrations (< 0.5 mg N/L), which is in the range of the nitrogen affinity index for NOB *Nitrospira* (K_s-NO_2^- = 0.13-0.38 mg N/L) (Nowka 2014).

With the constant low oxygen concentrations that were applied, larger (denser) aggregates will have an increased anoxic zone due to mass transfer limitations, resulting in an increased competition for nitrite between between denitrifiers, AnAOB and NOB, and thus increased suppression of NOB in the system (Vlaeminck *et al.* 2010; Volcke *et al.* 2012). This hypothesis can be confirmed with the measured R-max values of the AerAOB, which increased faster compared to those of NOB, proving enrichment of AerAOB vs. NOB, while being positively correlated with the average particle size in the reactor.

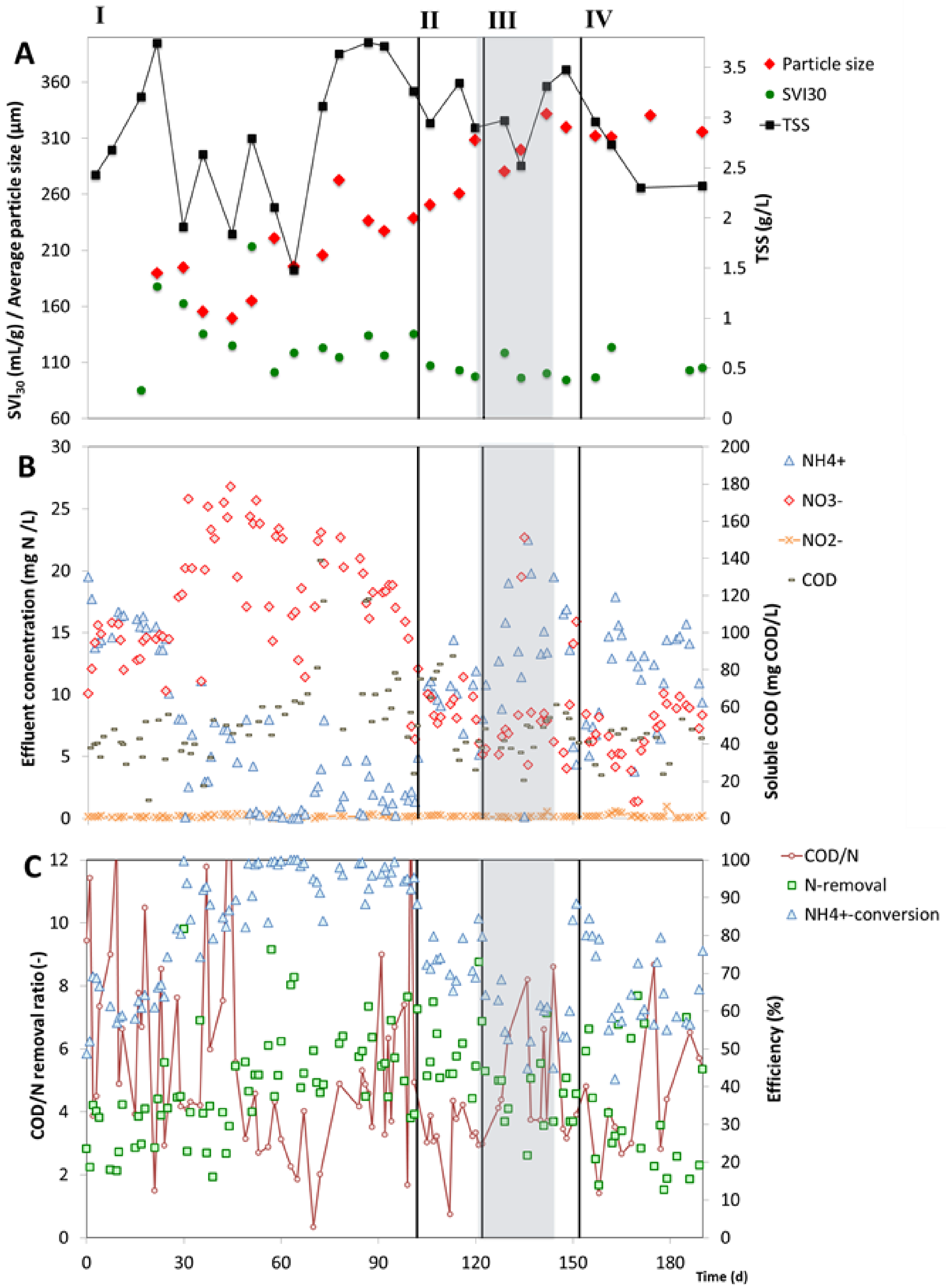


Figure 2. Performance of the reactor. A) Sludge characteristics, B) Effluent concentrations, C) Nitrogen and COD conversion. The grey-blue area indicates a period with seeding from a sidestream PN/A reactor.

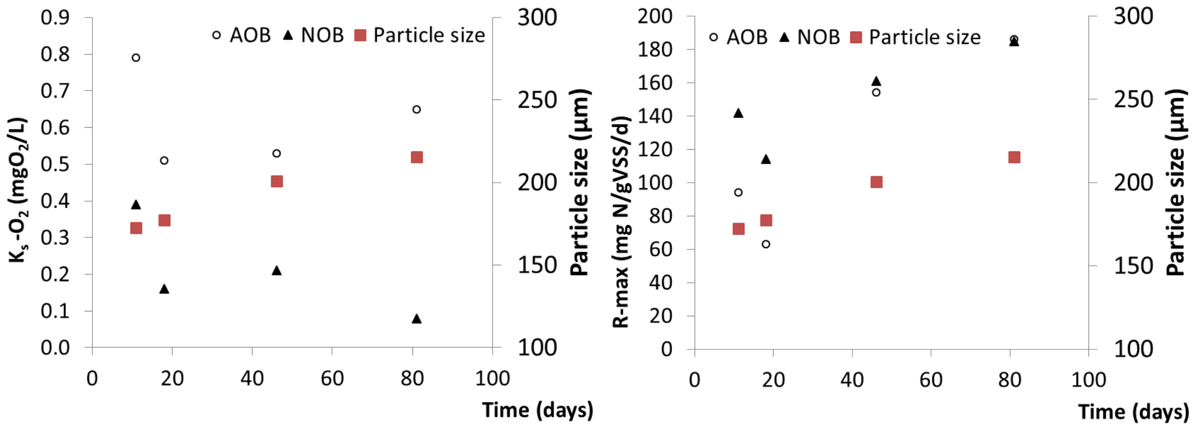


Figure 3. Measured oxygen affinity indices (K_s-O_2), maximum removal rates (R-max) and average particle size $Ø_{4,3}$ during period I

Furthermore, several authors studied the effect of mass transfer limitations on nitrifying kinetics in batch tests (Beccari *et al.* 1992; Manser *et al.* 2005; Blackburne *et al.* 2007). They concluded that higher particle sizes resulted into mass transfer limitations for oxygen, starting from a particle size of 40 µm, resulting in increased apparent Ks-O_2 and decreased R-max values. The apparent values obtained in this long term experiment show however increased R-max rates and no significant influences on the Ks-O_2 values, indicating adaptation of both AerAOB and NOB to the operational conditions. From this it can be concluded that competition for nitrite in the anoxic zone, rather than mass transfer limitations, will have played the main role in suppression of NOB in the system.

Effect of shear

During the first month of operation, different upflow velocities were tested. While increasing the upflow velocity to 2.3 m/h (top) and 8.4 m/h (bottom), the amount of shear conveyed by pumping increased. Due to the application of increased shear on the sludge from day 16 to day 50, a decrease in average particle size was measured from 190 to 140 µm. This resulted in a steady-state N-removal during this period. From day 51, an upflow velocity in the reactor of 2.3 m/h (bottom) and 0.45 m/h (top) was applied, avoiding disruption of the freshly formed aggregates.

Effect of pH

The pH in the reactor at the start of the experiment was low due to the influent spiking with an acidic solution of ammonium sulfate. With the addition of a pH control on day 38, the pH was increased from 6.5 to 6.8. This change in conditions increased the AerAOB activity in the reactor, and the ammonium conversion increased from 80 to 100%. No effect on the amount of nitrogen removal was measured. This increase in AerAOB activity is in line with earlier reported pH sensitivity for nitrifiers (Henze 2008).

Period II

In a second period, an extra selection phase was introduced in the reactor cycle to increase further granulation pressure. This selection phase consists of a mixing step with aeration, followed by a settling time that defined the MSV of the biomass (2.5-3 m/h). The frequency of this step could also be chosen. At the same moment, the nitrogen loading of the reactor was increased and the aeration time was decreased to ensure a residual amount of ammonium in the reactor to render the AerAOB a kinetic advantage over the NOB.

Overall performance

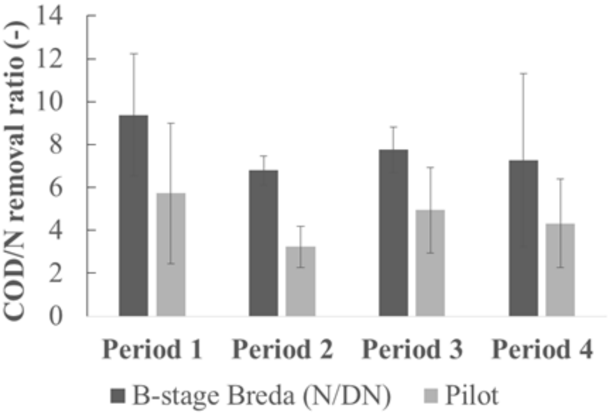
The extra selection pressure resulted in an increased particle size distribution, with an average size of $\emptyset_{4,3}$ = 280 µm. The 10% largest particles, $d_{0.9}$, were larger than 600 µm, indicating the formation of larger aggregates within the sludge, whereas the 10% smallest particles ($d_{0.1}$) were smaller than 100 µm, pointing out the wash-out of smaller flocs in the system. The SVI_{30} values affirm a well settling sludge (SVI_{30} < 100 mL/g) that can be used for a continuously fed process, where the sludge settles below the discharge point during the anoxic phase, and thus no external settlers or settling phase are needed anymore. With further increase of particle size, no further increase in NOB suppression in the reactor was obtained.

Similar COD/N removal ratios (3.2±0.9) were present during the operation, indicating the presence of the nitrite shortcut and with 51±23% removal of nitrogen over nitrite. To compare these COD/N removal ratios, the COD/N removal ratios of the B-stage in Breda, that uses the same influent as the pilot and where conventional N/DN is utilized, were calculated (Figure 4). The calculated values indicate a higher ratio than achieved by the process, showing further proof of the efficient use of carbon for nitrogen removal in the reactor. Judging effluent concentrations, nitrogen concentrations were still too high to be discharged (TN > 10 mg N/L), yet COD effluent levels were reached (<100 mg COD/L) and slightly higher than effluent COD levels of the B-stage (34±3 mg COD/L).

The nitrogen removal efficiency during this period was similar than reported by Lotti *et al.* (2014a), 52±16% vs. 46±13%, operating a granular PN/A at similar temperature (19°C) and influent N-concentrations, yet lower specific removal rates were achieved (24 mg N/g VSS/d vs. 48 mg N/g VSS/d). This can be attributed to the presence of a more specialized, in terms of enriched AnAOB, granular community, seeded from sidestream conditions, that is able to remove higher loading rates. This type of seeding sludge was not present in our reactor, resulting in lower specific removal rates.

In a recent pilot study on Nit/DNit, Regmi *et al.* (2014) utilized some of the previous mentioned strategies in the introduction (**1,2,3 and 4**) to suppress NOB and reported a nitrite accumulation of 1-2 mg N/L on pretreated sewage, yet high COD/Total Inorganic Nitrogen (TIN) removal ratios in the range of 11-20 were reported (TIN/COD = 0.05-0.09). Compared to these ratios, our results show a lower carbon need to remove the nitrogen, COD/N = 3.2±0.96, yet lower removal efficiencies (52 vs. 80%). This can be attributed to the lower DO-setpoint that was used, 0.3-0.4 vs.1.6 mg O_2/L, decreasing oxidation of COD with O_2. The results show that an effective strategy of suppression of NOB has to go hand in hand with effective usage of carbon in the system.

Figure 4. COD/N removal ratios full scale nitrification/denitrification (N/DN) B-stage Breda vs. pilot.



Periods III & IV

In the next periods, the frequency of the selection phase was further increased, first a factor 3 higher to 3/24 cycles and then a factor 1.33 higher to 4/24 cycles, with a selection pressure of 3 m/h. In period IV, the aerobic time was decreased from 45 to 39%.

Overall performance

The application of the extra selection stress on the reactor led to washout of AerAOB in the reactor, combined with a decrease of the amount of sludge in the reactor in period IV. This led to a decrease in nitrogen removal from 34% in period III to 23% in period IV. The reason for the increase in NOB activity at the end of period IV is unclear, because no further operational changes were made, adaptation could be the reason.

The results indicate that the combination of a low DO concentration, increase in particle size, combined with the presence of high selection pressure for well settling sludge, resulted in the washout of AerAOB of the reactor. Strong SRT control should therefore always be accompanied by sufficient control of DO concentration and aeration time to ensure AerAOB presence.

Particle size distribution & effect of seeding

During period III, the reactor was seeded every weekday from day 119-142 with 100 L suspended sludge from the sidestream PN/A treatment plant in Breda. This sludge had a $Ø_{4,3}$ = 215 µm ($d_{0.1}$=39 µm, $d_{0.9}$=480 µm).

Over this period, an increase in sludge concentration was measured, which can be attributed to the addition of the sidestream PN/A sludge (±0.8 g TSS/L), yet the increase in particle size could not only be assigned to the seeding. Since the average particle size distribution of the sludge was lower than the one present in the reactor (avg. reactor $Ø_{4,3}$ = 280 µm), this means that further granulation was occurring during the addition of the sidestream PN/A sludge. Furthermore, the semi-granular sludge was maintained during the end of period III and in period IV, indicating the formation of aggregates of this size ($d_{0.9}$ = 600 µm) in the reactor. Yet, the addition of this sludge could have partly contributed to the increase in particle size in the reactor, since no further increase was measured after the seeding stopped. Larger particles could have been retained in the reactor, whereas smaller would have been washed out.

The seeding of the sidestream PN/A sludge could not have influenced the measured nitrogen balance, since it contained only 3% of the total daily N-load (100 L/d, 239 mg N/L). Furthermore, no significant effect on the nitrogen balance was observed, suggesting that addition of AnAOB and AerAOB to the reactor did not improve its nitrogen removal. This is also reflected in the COD/N removal ratios during period III, which were higher than in period II. The limitation of AerAOB activity, which was already present due to operational conditions prior to seeding, resulted in no extra nitrite production, and thus substrate limitation for AnAOB, limiting growth and activity.

This lower AerAOB activity is in line with reported results by Lotti *et al.* (2014a), operating a granular system, while imposing an aggressive SRT on the flocculant biomass at lower temperatures and DO concentrations (20-30°C). Yet, these lower DO setpoint levels are necessary to withhold competition for nitrite between the anoxic organisms and NOB in the system. Other strategies use higher DO setpoints and aggressive SRT to outcompete NOB, but may inhibit AnAOB while exposing it to higher oxygen concentrations and lower temperatures**.**

CONCLUSIONS

A combination of different kinetic NOB suppression and sludge selection strategies is necessary to outcompete NOB under mainstream wastewater conditions. This study made use of kinetic selection strategies for specific stimulation and suppression of certain microbes, like the nitratational lag, residual NH_4^+ levels, and creating anoxic space for nitrite consumption, in combination with increased sludge settling pressure, leading to the development of a semi-granular sludge that partly suppressed NOB. This resulted in a sludge that was able to remove nitrogen partly over nitrite, with low COD/N removal ratios of 3.2±0.9 and nitrogen removal efficiencies of 52±16%. These COD/N removal ratios are lower than the current operation of the full-scale B-stage in Breda (6.8-9.4), and show promising results for carbon efficient nitrogen removal over nitrite, while producing a well settling sludge (SVI_{30}<100 mL/g).

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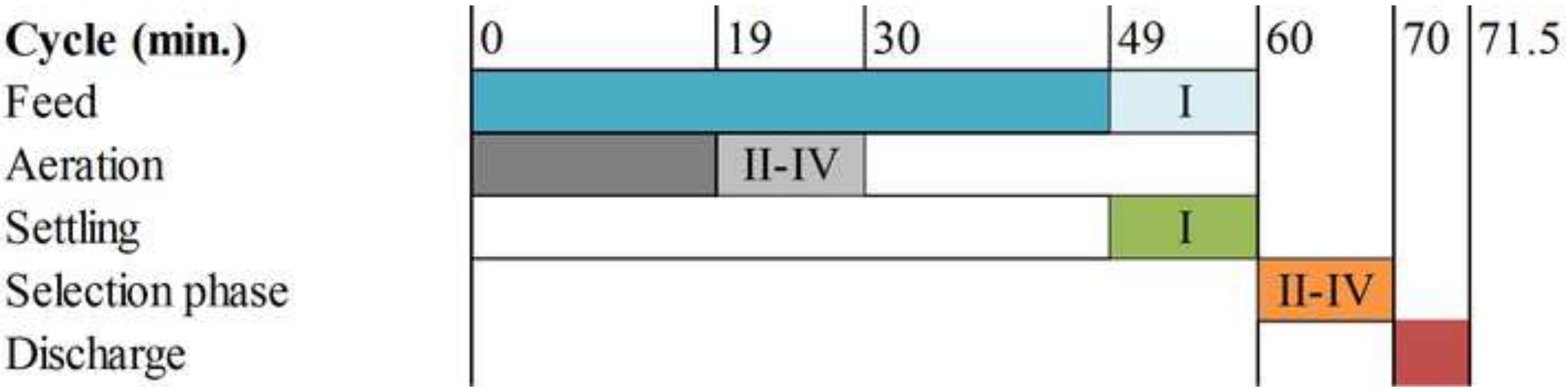
Figure 1


Cycle (min.)
Feed
Aeration
Settling
Selection phase
Discharge
0
19
30
49
60
70
71.5
I
II-IV
I
II-IV

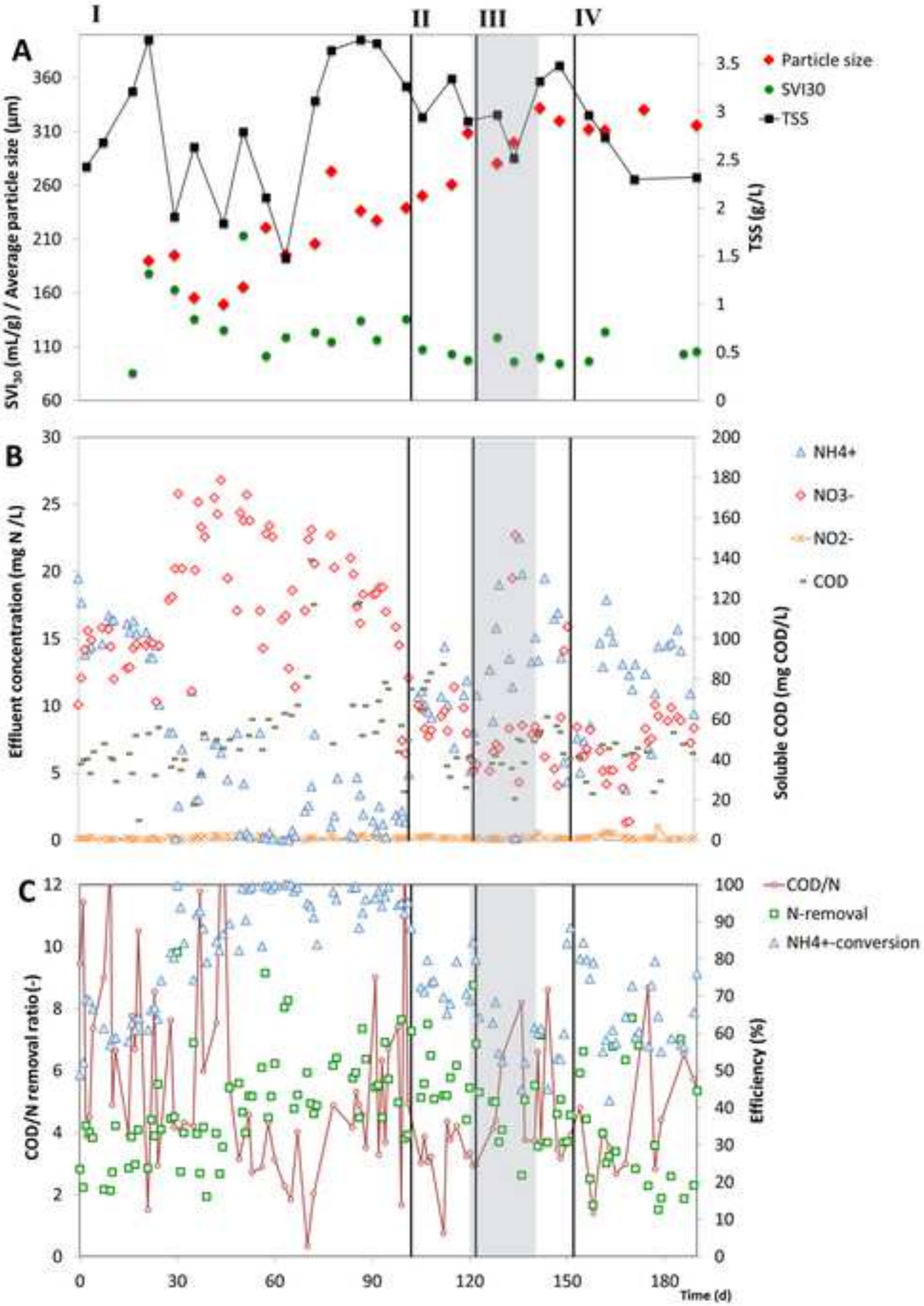
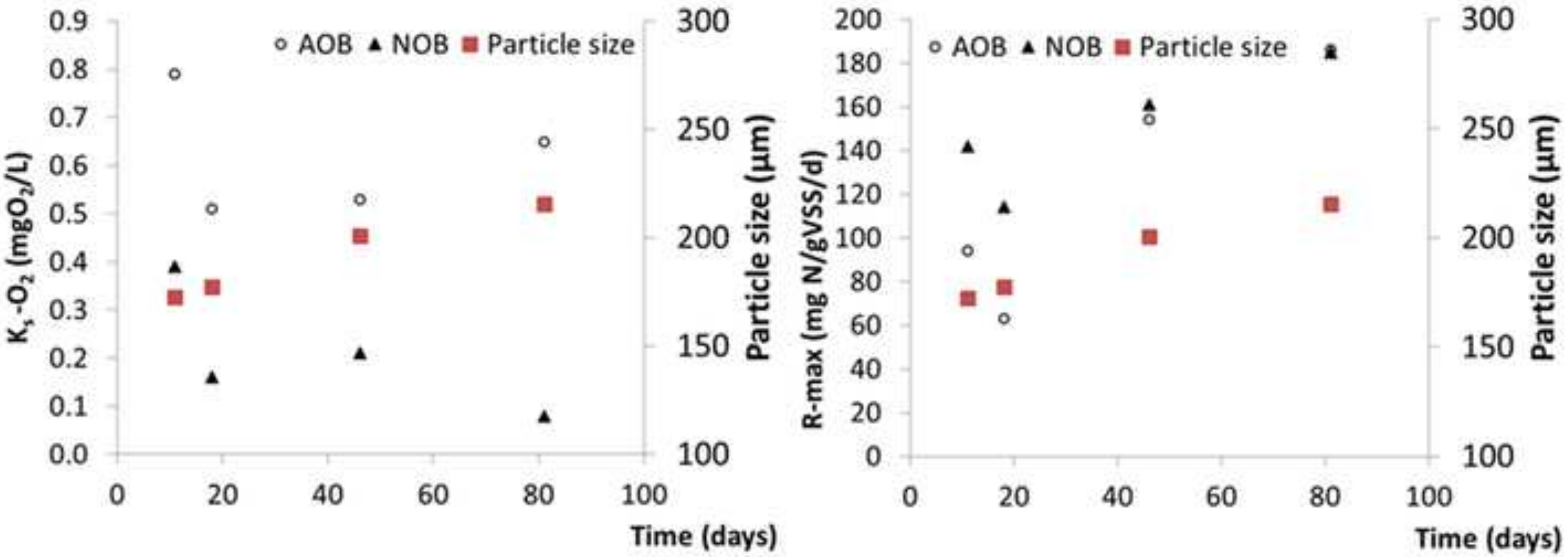

I
II
III
IV
A
SVI$_{30}$ (mL/g) / Average particle size (µm)
360
310
260
210
160
110
60
TSS (g/L)
3.5
3
2.5
2
1.5
1
0.5
0
Particle size
SVI30
TSS
B
Effluent concentration (mg N /L)
30
25
20
15
10
5
0
Soluble COD (mg COD/L)
200
180
160
140
120
100
80
60
40
20
0
NH4+
NO3-
NO2-
COD
C
COD/N removal ratio (-)
12
10
8
6
4
2
0
Efficiency (%)
100
90
80
70
60
50
40
30
20
10
0
COD/N
N-removal
NH4+-conversion
0
30
60
90
120
150
180
Time (d)

Figure 3



0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0.0
K_s-O_2 (mgO$_2$/L)
○ AOB ▲ NOB ■ Particle size
300
250
200
150
100
Particle size (µm)
0
20
40
60
80
100
Time (days)
200
180
160
140
120
100
80
60
40
20
0
R-max (mg N/gVSS/d)
○ AOB ▲ NOB ■ Particle size
300
250
200
150
100
Particle size (µm)
0
20
40
60
80
100
Time (days)

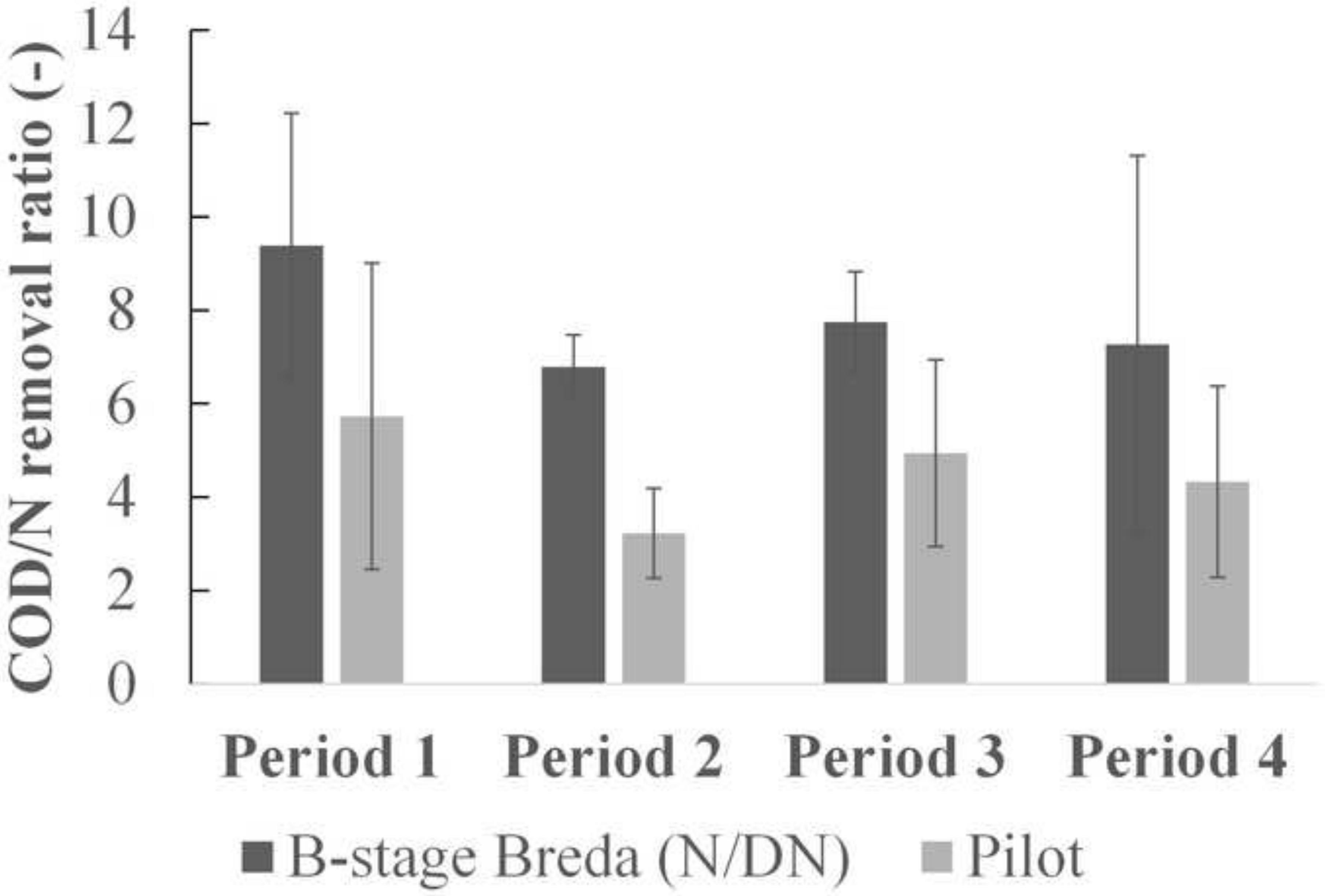
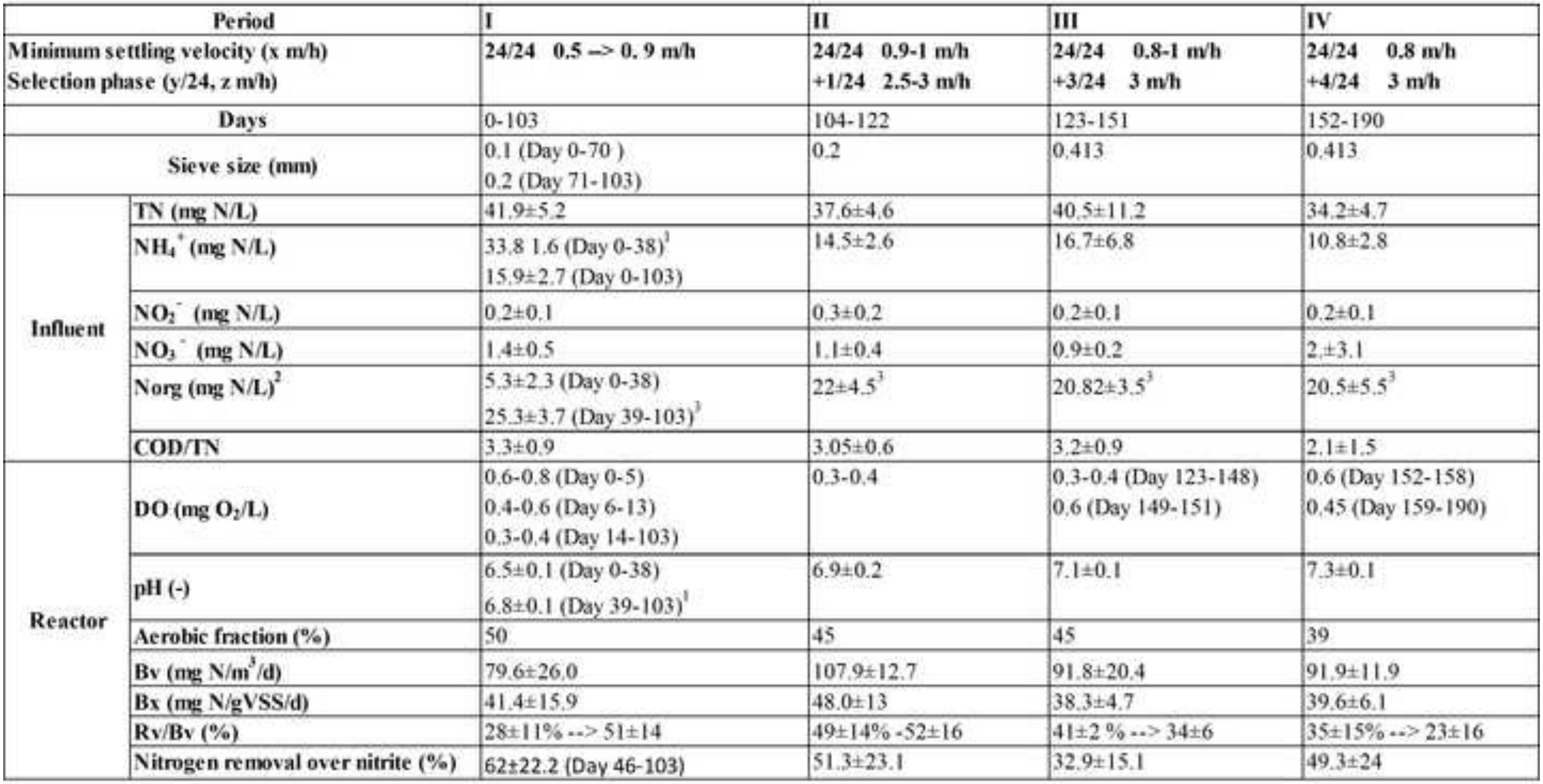

COD/N removal ratio (-)
0
2
4
6
8
10
12
14
Period 1
Period 2
Period 3
Period 4
B-stage Breda (N/DN)
Pilot

Table 1



Period		I	II	III	IV
Minimum settling velocity (x m/h) Selection phase (y/24, z m/h)		**24/24 0.5 --> 0. 9 m/h**	**24/24 0.9-1 m/h +1/24 2.5-3 m/h**	**24/24 0.8-1 m/h +3/24 3 m/h**	**24/24 0.8 m/h +4/24 3 m/h**
Days		0-103	104-122	123-151	152-190
Sieve size (mm)		0.1 (Day 0-70) 0.2 (Day 71-103)	0.2	0.413	0.413
Influent	**TN (mg N/L)**	41.9±5.2	37.6±4.6	40.5±11.2	34.2±4.7
	NH_4^+ (mg N/L)	33.8 1.6 (Day 0-38)[1] 15.9±2.7 (Day 0-103)	14.5±2.6	16.7±6.8	10.8±2.8
	NO_2^- (mg N/L)	0.2±0.1	0.3±0.2	0.2±0.1	0.2±0.1
	NO_3^- (mg N/L)	1.4±0.5	1.1±0.4	0.9±0.2	2.±3.1
	Norg (mg N/L)[2]	5.3±2.3 (Day 0-38) 25.3±3.7 (Day 39-103)[3]	22±4.5[3]	20.82±3.5[3]	20.5±5.5[3]
	COD/TN	3.3±0.9	3.05±0.6	3.2±0.9	2.1±1.5
Reactor	**DO (mg O_2/L)**	0.6-0.8 (Day 0-5) 0.4-0.6 (Day 6-13) 0.3-0.4 (Day 14-103)	0.3-0.4	0.3-0.4 (Day 123-148) 0.6 (Day 149-151)	0.6 (Day 152-158) 0.45 (Day 159-190)
	pH (-)	6.5±0.1 (Day 0-38) 6.8±0.1 (Day 39-103)[1]	6.9±0.2	7.1±0.1	7.3±0.1
	Aerobic fraction (%)	50	45	45	39
	Bv (mg N/m^3/d)	79.6±26.0	107.9±12.7	91.8±20.4	91.9±11.9
	Bx (mg N/gVSS/d)	41.4±15.9	48.0±13	38.3±4.7	39.6±6.1
	Rv/Bv (%)	28±11% --> 51±14	49±14% -52±16	41±2 % --> 34±6	35±15% --> 23±16
	Nitrogen removal over nitrite (%)	62±22.2 (Day 46-103)	51.3±23.1	32.9±15.1	49.3±24