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Optimization of a mainstream nitrification-denitrification process and anammox polishing

Pusker Regmi, Becky Holgate, Dana Fredericks, Mark W. Miller, Bernhard Wett, Sudhir Murthy and Charles B. Bott

ABSTRACT

This paper deals with an almost 1-year long pilot study of a nitrification-denitrification process that was followed by anammox polishing. The pilot plant treated real municipal wastewater at ambient temperatures. The effluent of high-rate activated sludge process (hydraulic retention time, HRT = 30 min, solids retention time = 0.25 d) was fed to the pilot plant described in this paper, where a constant temperature of 23 °C was maintained. The nitrification-denitrification process was operated to promote nitrite oxidizing bacteria out-selection in an intermittently aerated reactor. The intermittent aeration pattern was controlled using a strategy based on effluent ammonia and nitrate + nitrite concentrations. The unique feature of this aeration control was that fixed dissolved oxygen set-point was used and the length of aerobic and anoxic durations were changed based on the effluent ammonia and nitrate + nitrite concentrations. The anaerobic ammonia oxidation (anammox) bacteria were adapted in mainstream conditions by allowing the growth on the moving bed bioreactor plastic media in a fully anoxic reactor. The total inorganic nitrogen (TIN) removal performance of the entire system was $75 \pm 15\%$ during the study at a modest influent chemical oxygen demand (COD)/NH₄⁺-N ratio of 8.9 ± 1.8 within the HRT range of 3.1–9.4 h. Anammox polishing contributed 11% of overall TIN removal. Therefore, this pilot-scale study demonstrates that application of the proposed nitrification-denitrification system followed by anammox polishing is capable of relatively high nitrogen removal without supplemental carbon and alkalinity at a low HRT.

Key words | AvN, anammox, aeration control, moving bed biofilm reactor (MBBR)

INTRODUCTION

Wastewater treatment plants (WWTPs) around the world are facing technical and financial challenges to meet ever more stringent water quality standards. For the WWTPs required to remove nitrogen, the cost of energy and resources (e.g., external carbon and alkalinity) for nitrogen removal from wastewater is increasing while nitrogen limits are becoming lower. Recently, there has been an explosion of new innovative technologies to achieve high levels of total nitrogen removal for less energy, fewer resource demands, and across less space. However, most of these technologies are limited to nitrogen-rich waste streams with low carbon to nitrogen ratio (COD/N hereafter). The very efficient partial nitrification-anaerobic ammonia oxidation (anammox) or deammonification-based technologies have already been

proven to treat high ammonia strength reject water with more than 100 full-scale installations (Lackner *et al.* 2014), while the mainstream implementation is currently under development by several research groups around the world. The latter holds the key to intensification of wastewater treatment for biological nitrogen removal (BNR), recovery of energy from carbon content of raw wastewater and the minimization of energy and resources for nitrogen polishing to meet stringent permits.

Successful mainstream deammonification depends on stable anammox activity, however, effectively suppressing nitrite oxidation is the major precondition. The most common approaches to suppress nitrite oxidizing bacteria (NOB) are specific to unique conditions of high temperature

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and high ammonia strength waste streams such as the reject stream generated from anaerobic digestion of municipal sludge. The implementation of partial nitrification + anammox is successfully applied in highly loaded sidestream processes that use one or more of the following conditions to out-select NOB: high temperature (Hellings *et al.* 1998); low dissolved oxygen (DO) (Joss *et al.* 2009); low solids retention time (SRT) (van Dongen *et al.* 2001); and free ammonia (FA) inhibition (Anthonisen *et al.* 1976) and free nitrous acid (FNA) (Zhou *et al.* 2011). However, since FA inhibition is not available at the lower total ammonia concentrations that occur in domestic wastewater and temperatures vary enough so that reliable high temperature selection is not viable, different strategies are needed for NOB out-selection in mainstream processes. In a preliminary study using a single continuous-flow stirred-tank reactor (CSTR) with continuous flow, Regmi *et al.* (2014) demonstrated unique mainstream strategies for achieving sustained NOB out-selection (Table 1).

These strategies, however, have not yet been tested for reliable NOB out-selection in a wide range of loading conditions in more practical plug-flow configurations.

The slower doubling times and sensitivity of anammox bacteria (AMX) toward dissolved oxygen and high carbon concentrations were thought to be the obstacles for mainstream implementation. In a preliminary study, the mainstream anammox polishing moving bed biofilm reactor (MBBR) coupled with the ammonium oxidizing bacteria (AOB) vs. NOB process (AvN) CSTR demonstrated a highly stable mainstream deammonification process (Regmi *et al.* 2015). The limitation of NOB out-selection in the AvN reactor resulting in more NO_3^- -N than NO_2^- -N severely limited the nitrogen turnover through the anammox metabolism.

Recently, the capability of certain AMX species to oxidize volatile fatty acids with NO_3^- as the electron acceptor has been demonstrated (Gueven *et al.* 2005, Kartal *et al.* 2007, Winkler *et al.* 2012). Since AMX converts the fatty acids directly to CO_2 without incorporating the volatile fatty acids (VFAs) into biomass, the yield associated with such metabolism is low (Winkler *et al.* 2012) and can be considered advantageous. Conversely, at a carbon to nitrogen ratio greater than 1, heterotrophs are shown to out-compete AMX (Gueven *et al.* 2005). The removal of NO_3^- produced from the AMX reaction or resulting from uninhibited NOB is of significant importance when anammox is used for nitrogen polishing to meet stringent permits.

In this study, an A-B process was piloted with the principal objective of repressing nitrite oxidation and implementing of anammox in mainstream. The A-stage (i.e., the high-rate carbon removal process) was operated under strategies to control the effluent carbon to ammonia ratio ($8 > \text{COD}/\text{NH}_4^+\text{-N} < 11$) that would be optimum for the B-stage for nitrogen removal. The B-stage, the focus of this paper, consisted of an aggressively operated (i.e., short SRT and HRT) nitrification-denitrification process (named AvN) and was followed by an anammox MBBR for nitrogen polishing. The combined AvN and the anammox MBBR was named AvN+. The B-stage AvN was operated under an intermittent aeration control strategy that targeted an effluent oxidized nitrogen to ammonia nitrogen $[(\text{NO}_2^- \text{-N} + \text{NO}_3^- \text{-N})/\text{NH}_4^+ \text{-N}]$ ratio of 1. In this paper, two aspects of the pilot study are presented: (1) NOB out-selection in AvN and nitrogen removal performance by the AvN+ process at different loading and operating conditions; and (2) the enhancement of nitrate and ammonia removal in the anammox MBBR with acetate ($\text{COD}/\text{NO}_3^- \text{-N} < 1.5$) addition to meet stringent nitrogen permit limits.

Table 1 | Strategies used by Regmi *et al.* (2014) to achieve NOB out-selection during mainstream treatment

Strategy	Action	Impact	Control basis
1	$\frac{\text{NH}_4^+}{\text{NO}_2^- + \text{NO}_3^-} = 1$	Optimum aerated and unaerated volume for nitrification and denitrification. Optimum alkalinity for AOB growth. Residual ammonium supports higher AOB growth rates	Control based on real-time effluent NH_4^+ , NO_2^- , NO_3^- signals
2	Intermittent aeration and bioavailable COD	Allow NO_2^- consumption by denitrifiers	Upstream organic carbon treatment system
3	Residual NH_4^+ -N	Allows AOB to operate at maximum rate	Control based on real-time effluent NH_4^+ , NO_2^- , NO_3^- signals
4	DO = 1.6 mg/L	AOBs grow faster than NOBs	Control DO set-point
5	Aggressive low SRT	AOBs to grow rapidly, making them competitive relative to stressed NOBs due to strategies 1, 2 and 3	Wasting

MATERIAL AND METHODS

The AvN+ pilot

The pilot process described in this study was part of a larger configuration including a high-rate activated sludge A-stage (HRT = 30 min, SRT = 0.25 days) for COD removal providing the influent for the AvN reactor. In this paper, the focus is only on the performance and operation of the AvN+ process (Figure 1). The AvN process was seeded with non-nitrifying biomass from a high-rate activated

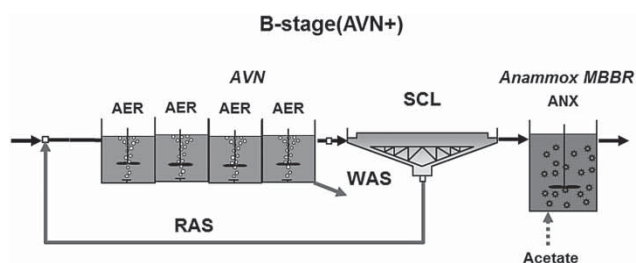


Figure 1 | Process flow diagram of the AvN+ process.

sludge plant, while the anammox MBBR was enriched with AMX on the biofilm carriers from a prior study (Regmi *et al.* 2015).

The AvN process included four equal sized reactors with combined aeration tank volume of 0.6 m^3 followed by a clarifier. Each reactor was equipped with a variable speed mixer ($G = 106 \text{ s}^{-1}$) in order to maintain completely-mixed conditions. Returned activated sludge from the clarifier was returned to the first AvN reactor with a peristaltic pump at 100% of the influent flow rate. SRT was controlled by wasting solids from the last reactor with a programmable digital peristaltic pump. All AvN reactors were equipped with sensors to monitor DO (Hach LDO, CO, USA) while the last reactor was also monitored for NO_3^- , NO_2^- (Spectrolyser, Austria), and NH_4^+ -N (WTW VARION, Germany). NH_4^+ -N, NO_3^- -N, NO_2^- -N signals were used to control aeration. The aeration control implemented has been described in detail by Regmi *et al.* (2015) (Figure 2).

The fully anoxic anammox MBBR had a volume of 0.45 m^3 where 50% of the volume was filled with K3 biofilm carriers (AnoxKaldnes, Sweden). The volume was later

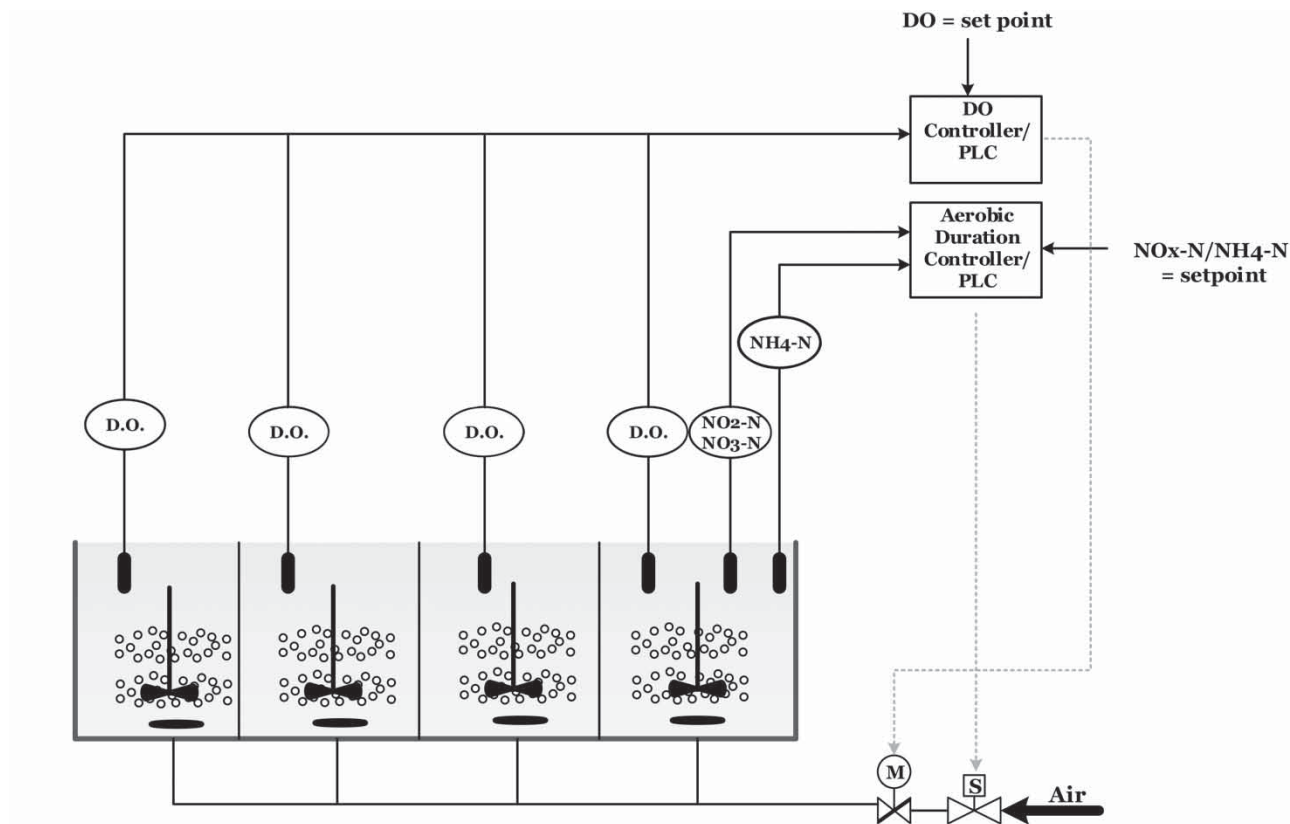


Figure 2 | AvN controller depicting aerobic duration controller receiving NH_4^+ , NO_2^- and NO_3^- signals and DO controller receiving dissolved oxygen signal.

changed to 0.34 m^3 on day 85. The effective surface area of the carriers was $500 \text{ m}^2/\text{m}^3$. Mechanical mixing of carriers was achieved by a variable speed mixer ($G = 14 \text{ s}^{-1}$). The pH was recorded continuously by an online pH probe and the reactor was covered with styrofoam to avoid oxygen transfer from the atmosphere. Acetate (sodium acetate) was fed to the anammox MBBR during days 101–191 and days 240–296.

To assess the performance of the AvN+ process within different loading conditions the HRT of the entire system was changed throughout the study.

Influent/effluent monitoring

Performance of the AvN pilot was monitored by collecting 24-h flow-weighted composite samples from the influent and effluent of the system. Samples were analyzed for total suspended solids (TSS) and volatile suspended solids (VSS), total and soluble chemical oxygen demand (COD), total Kjeldahl nitrogen (TKN), total phosphorus (TP), organophosphate (OP), NO_3^- -N, NO_2^- -N, NH_4^+ -N and alkalinity. Total inorganic nitrogen (TIN) is the sum of NO_2^- -N, NO_3^- -N, and NH_4^+ -N. Nitrite accumulation rate (NAR) is simply the ratio of NO_2^- -N to the sum of NO_3^- -N + NO_2^- -N.

Microbial activity measurements

To measure maximum AOB and NOB activity rates, 4 L samples were collected and dispensed into 4 L vessels from the AvN CSTR and aerated for 30 min to oxidize excess COD, and spiked with 20–30 mg/L NH_4^+ -N (as ammonium chloride) and 2–4 mg/L NO_2^- -N (as sodium nitrite), and sampled continuously for 1 h at 20-min intervals. All collected samples were analyzed for NH_4^+ -N, NO_2^- -N, and NO_3^- -N. Mixing was provided by magnetic stirring. The dissolved oxygen (DO) concentration was maintained between 2.5 and 4 mg O_2/L . pH was maintained between 7 and 7.5 by adding sodium bicarbonate. The AOB rates were calculated as the slope of the NO_x -N production and NOB rates were calculated as the slope of the NO_3^- -N production.

AMX activity tests

To measure maximum AMX activity, the anammox MBBR was isolated from the system. A sample was taken for soluble COD (sCOD), NH_4^+ -N, NO_2^- -N, and NO_3^- -N. The MBBR was spiked with 8 mg/L NH_4^+ -N (as ammonium

chloride) and 10 mg/L NO_2^- -N (as sodium nitrite) and sampled continuously at 20-min intervals until the NO_2^- -N was less than 1.5 mg/L NO_2^- -N. On the last sample of the activity measurement, a sCOD sample was taken along with NH_4^+ -N, NO_2^- -N, and NO_3^- -N. The DO concentration was maintained close to 0 mg O_2/L and was recorded at every 20 min interval along with the pH. NH_4^+ -N and NO_2^- -N uptake rates were calculated as the slope of the NH_4^+ -N and NO_2^- -N values taken during the activity test, respectively. NO_3^- -N production rates were calculated as the slope of the NO_3^- -N values. The results of the tests were presented in units of g N/ m^2/d .

Biomass density

Biomass density measurements were performed according to the method described by Regmi *et al.* (2011). Measurements were performed bi-weekly during the study.

Molecular methods for microbial quantification

DNA and RNA extraction was conducted using the DNeasy and RNeasy mini kits (Qiagen, CA). Resulting DNA and RNA concentrations and quality were initially checked by UV spectrophotometry (Varian, CA). The abundance of AOB and NOB were quantified via SYBR[®] Green chemistry quantitative polymerase chain reaction (qPCR) assays, NH_4^+ monooxygenase subunit A (*amoA*) gene (Rotthauwe *et al.* 1997), *Nitrobacter* 16S rRNA gene (Graham *et al.* 2007) and *Nitrospira* 16S rRNA gene (Kindaichi *et al.* 2007), respectively. Total bacterial abundance was quantified using eubacterial 16S rRNA gene targeted primers (Ferris *et al.* 1996). qPCR assays were conducted on a iQ5 real-time PCR thermal cycler (BioRad Laboratories, Hercules, CA). Standard curves for qPCR were generated via serial decimal dilutions of plasmid DNA containing specific target gene inserts. qPCR for standard plasmid DNA and sample DNA were conducted with duplication and triplication, respectively. DNA grade deionized distilled water (Fisher Scientific, MA) was used for non-template control. Primer specificity and the absence of primer-dimers were confirmed via melt curve analysis of each and every qPCR profile.

The abundance of AMX was quantified via SYBR[®] Green chemistry quantitative PCR (qPCR) assays targeting AMX 16S rRNA gene (van der Star *et al.* 2007). *C. Brocadia fulgida* specific qPCR assay was applied based on the highly variable region of the *hzsA* gene. qPCR primers were used with TaqMan chemistry (forward, 5'-AGT TAG TGA GTG

TGG ATG GCG TGT-3'; reverse, 5'-TCA TCC TGC GTG AGG AAC TTG TCA-3'; probe, 5'-/56-FAM/AT TCA GCC G/Zen/T GCG TAC ACC AGC TTG CTT /3IABkFQ/-3') (IDTDNA, IA).

qPCR assays were conducted on a iQ5 real-time PCR thermal cycler (BioRad Laboratories, CA). Standard curves for qPCR were generated via serial decimal dilutions of plasmid DNA containing specific target gene inserts. qPCR for standard plasmid DNA and sample DNA were conducted with duplication and triplication, respectively. DNA grade ddH₂O (Fisher Scientific, MA) was used for non-template control. Primer specificity and the absence of primer dimers were confirmed via melt curve analysis.

RESULTS AND DISCUSSION

AvN+ nitrogen removal performance

AOB activity in the AvN process was established within two weeks into the study with the effluent NH₄⁺-N below 10 mg N/L (Figure 3(a)). NOB activity was lower than AOB activity during this time as high concentrations of NO₂⁻-N were observed in the effluent. The anammox MBBR removed NO₂⁻-N and NH₄⁺-N without any indication of lag during the startup (see later, Figure 5(b)). The influent NH₄⁺-N concentration to the AvN system fluctuated between 25 and 40 mg N/L (Figure 3(a)).

The key effluent parameters of the A-stage (influent to the AvN), AvN and anammox MBBR averaged over the 296 days of operation are presented in Table 2. The average influent COD/NH₄⁺-N ratio to the AvN was 8.9, which can be considered limiting to achieve a high degree of nitrogen removal in a combined carbon and nitrogen removal system through nitrification and denitrification (Grady et al. 2011). Almost half of the influent COD to the AvN was in the soluble form. The AvN effluent NH₄⁺-N averaged 6.6 and the average NO_x-N was 6.7 during the entire study, which demonstrates that the AvN aeration controller was able to achieve its goal.

The TIN removal performance of the AvN+ system was 75 ± 15% during the study at a modest influent COD/NH₄⁺-N ratio of 8.9 ± 1.8. In Phase III (AvN+ HRT = 9.1 h) the TIN removal efficiency averaged 91%, which was the highest among all phases. In Phase III the A-stage COD removal efficiency averaged 41% which resulted in the influent COD/NH₄⁺-N ratio of 10.5 (Figure 4).

The relative TIN removal contribution of the anammox MBBR was very stable within the study period. Since a significant amount of TIN removal was taking place in the AvN process during Phase III, the anammox contribution was limited to 8% which is lower than the average of 11% for the entire study. It is worthwhile to note that Phase III was also the period with the lowest NOB out-selection as indicated by nitrite accumulation ratio (NAR) of 0.16 and the ratio of maximum NOB activity and maximum AOB activity (NOB rate/AOB rate) of 1 (Table 3).

In Phases I, IV and V, AvN+ was operated at a very low HRT (Table 4). The overall TIN removal efficiency during these periods were similar, however, relatively high TIN removal contribution by anammox was observed. The high TIN removal contribution from anammox was due to relatively high NAR in the AvN effluent during those periods (Table 3). Consequently, it shows that AvN+ can be operated within a small volume to achieve relatively high nitrogen removal performance even with an aggressively operated upstream COD recovery system. The key to such performance was NOB out-selection in the AvN process, which provided effluent with high concentrations of NO₂⁻-N for significant nitrogen polishing via anammox metabolism.

Avn NOB out-selection and overall performance

The TIN removal performance of the AvN process was stable during the study and was mostly dependent on the influent COD/NH₄⁺-N ratio (Table 3). The NH₄⁺-N loading rate to the AvN process was varied by changing the HRT of the system during different phases of the study. During Phase IV, the average NH₄⁺-N loading rate was 339 mg N/L/d with average TIN removal rate and TIN removal efficiency of 172 mg N/L/d and 51%, respectively, at an average influent COD/NH₄⁺-N ratio of 9.1 (Table 3).

The relatively low average TIN removal efficiency during Phase IV could be explained by a sudden loss of mixed liquor suspended solids (MLSS) on day 178 due to a clarifier malfunction (Figure 5). The consequence of rapid loss of AOB population during Phase IV was the increase in the aerobic fraction (which is controlled to maintain effluent NH₄⁺-N = effluent NO_x-N), which causes more aerobic oxidation of COD, and less COD being available for NO_x-N reduction (Table 3). The result of this is reflected in a high ratio of COD removal rate and TIN removal rate (14.0 ± 3.1) during Phase IV (Table 3). Similarly, in Phase

Figure 5 | Trends of key parameters for the assessment of NOB out-selection: (a) NH_4^+ -N loading rate and the ratio of maximum NOB rate to maximum AOB rate; (b) NH_4^+ -N loading rate and AvN aerobic fraction; (c) MLSS and aerobic SRT. Note: There was a sudden drop in mixed liquor due to clarifier malfunction on days 62 and 178 represented by a gray shaded area on the graph.

maximum activity measurements were close to theoretical values proposed by Strous *et al.* (1998). The biomass density increased steadily during the course of the study reaching 5.8 g/m^2 from the initial biomass density of 2.7 g/m^2 (Table 5). Although the NO_2^- -N loading was decreased during Phases II and III, the biomass density continued to increase (Table 5).

Figure 6 | Trends of microbial populations (AOB, NOB and total bacteria) presented as copies of DNA per mL of sample from targeted qPCR (a) and weekly AOB and NOB maximum rate measurements (b).

With the goal of improving TIN removal efficiency, a limited amount of acetate was added to the anammox MBBR from days 102 to 161 and 241 to 296. During Phase III, the TIN removal efficiency averaged 50%, which was highest among all phases despite receiving the lowest amount of NO_2^- -N (Table 5). This was possible due to the addition of acetate which was responsible for NO_3^- -N removal (Table 6). In fact, the NO_3^- -N removal during the acetate addition was 2–3 times more than NO_3^- -N removal than was observed without acetate addition (Table 6). Further, the NH_4^+ -N removal in the anammox MBBR during acetate addition was much higher than could be accounted for by the anammox stoichiometric value (NO_2^- -N removed: NH_4^+ -N removed = 1.32). The ratio of NO_2^- -N removed: NH_4^+ -N removed was lower during the period of acetate addition (Table 6). On the contrary the ratio of NO_x -N removed: NH_4^+ -N removed was closer

Table 5 | Performance and other relevant data of the anammox MBBR during the study period of 296 days (Average \pm standard deviation)

Parameter	Phase I	Phase II	Phase III	Phase IV	Phase V	Phase VI
TIN removal rate (g N/m ² /d)	0.23 \pm 0.08	0.11 \pm 0.04	0.09 \pm 0.06	0.36 \pm 0.07	0.27 \pm 0.05	0.14 \pm 0.03
NO ₂ ⁻ -N loading rate (g N/m ² /d)	0.118 \pm 0.058	0.038 \pm 0.021	0.012 \pm 0.004	0.182 \pm 0.051	0.131 \pm 0.032	0.044 \pm 0.010
TIN removal efficiency (%)	33 \pm 9	35 \pm 13	50 \pm 17	38 \pm 7	47 \pm 12	46 \pm 11
Max TIN removal rate (g N/m ² /d)*	0.71 \pm 0.06	1.35 \pm 0.16	1.03 \pm 0.09	1.17 \pm 0.20	1.07 \pm 0.15	0.96 \pm 0.15
NO ₂ ⁻ -N removed/NH ₄ ⁺ -N removed*	1.47 \pm 0.20	1.42 \pm 0.02	1.45 \pm 0.14	1.49 \pm 0.13	1.45 \pm 0.12	1.59 \pm 0.10
NO ₃ ⁻ -N produced/NH ₄ ⁺ -N removed*	0.29 \pm 0.06	0.26 \pm 0.02	0.23 \pm 0.04	0.22 \pm 0.06	0.32 \pm 0.12	0.32 \pm 0.06
Biomass density (g/m ²)**	2.7 \pm 0.33	3.3 \pm 0.21	3.7 \pm 1.1	5.1 \pm 2.0	5.4 \pm 1.0	5.86 \pm 0.23

*From weekly *in-situ* AMX maximum activity measurements.

**From bi-weekly biomass density measurements of the MBBR media. The acetate was added to the anammox MBBR at the middle of Phase II and entire period during Phase III.

Table 6 | Assessment of anammox MBBR with and without acetate addition (Average \pm standard deviation)

	Day 1–101 (no acetate)	Day 102–161 (acetate added)	Days 162–240 (no acetate)	Days 241–296 (acetate added)
NO ₂ -N removed/NH ₄ -N removed	0.99 \pm 0.29	0.36 \pm 0.11	0.96 \pm 0.22	0.87 \pm 0.31
NO _x -N removed/NH ₄ -N removed	1.14 \pm 0.35	1.4 \pm 0.30	1.25 \pm 0.30	1.40 \pm 0.28
NO ₃ -N removed	0.32 \pm 0.52	1.25 \pm 0.38	0.61 \pm 0.40	1.1 \pm 0.60
Acetate COD/NO ₃ -N	0	0.91 \pm 0.60	0	0.96 \pm 0.20

to the anammox stoichiometric value (Table 6). This suggests that NO₃⁻-N reduction to NO₂⁻-N was supplying AMX their substrate for anaerobic NH₄⁺-N oxidation. In limited carbon addition (acetate) conditions certain species of AMX are known to reduce NO₃⁻-N to NO₂⁻-N to create their own substrate for NH₄⁺-N oxidation (Kartal *et al.* 2007). However, heterotrophic denitrification in carbon limited conditions could also provide NO₂⁻-N for anammox metabolism.

CONCLUSIONS

The AvN+ implemented downstream of a high-rate activated sludge process (HRT = 30 min) operated at an average COD removal efficiency of 50% demonstrated average TIN removal efficiency of 75% at 23 °C. When the AvN was operated aggressively (i.e., low HRT and short SRT) NOB out-selection was achieved. The *ex-situ* maximum activity tests revealed that the ratio of maximum NOB rate to maximum AOB rate was 0.75 \pm 0.17 during the study.

To meet stringent effluent nitrogen permits, energy and resource intensive tertiary treatment is used to eliminate the remaining few percentages of nitrogen, which accounts for the large carbon footprint in the entire plant (Falk

et al. 2013). In this study we demonstrated nitrogen polishing in a small footprint anammox MBBR without aeration and with little to no addition of external carbon sources (COD/NO₃⁻-N ratio of ~1). Nitrate production during the anammox reaction often limits the nitrogen removal through anammox metabolism. We showed removal of nitrate and thus induced ammonia removal by a post-polishing MBBR with anammox, which extends the applicability of anammox for mainstream nitrogen removal to meet lower effluent nitrogen permits at a limited external carbon dosage. The process intensification (i.e., reduction in physical footprint of the plant) and reduction in chemical usage to achieve advanced nitrogen removal will make the AvN+ process highly desirable in terms of economic feasibility of BNR.

Therefore, the AvN+ system exploiting short-cut nitrogen removal showed that COD capture for energy production is possible without compromising nitrogen removal performance.

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