Separation and Purification Technology 166 (2016) 102–108

Contents lists available at ScienceDirect

Separation and Purification Technology

journal homepage: www.elsevier.com/locate/seppur

Review

The presence of organic matter during autotrophic nitrogen removal: Problem or opportunity?

Elisa A. Giustinianovich [a], José-Luis Campos [b], Marlene D. Roeckel [a],*

[a] Department of Chemical Engineering, University of Concepción, Concepción, Chile
[b] Faculty of Engineering and Sciences, Universidad Adolfo Ibañez, Viña del Mar, Chile

ARTICLE INFO

Article history:
Received 17 August 2015
Received in revised form 18 March 2016
Accepted 10 April 2016
Available online 11 April 2016

Keywords:
Anammox
Ammonia oxidation
Heterotrophic denitrification
Nitrogen removal
SNAD

ABSTRACT

The simultaneous nitrification, Anammox and denitrification (SNAD) process discovered six years ago is an adaptation of the autotrophic denitrification process that allows for treating nitrogen-rich wastewater streams with moderate amounts of organic carbon. Several authors have noted that it is possible to utilize organic carbon to promote nitrogen removal via the action of denitrifying microorganisms, which can remove the remnant nitrate produced by Anammox bacteria. Thus, SNAD systems can achieve nitrogen removal efficiencies higher than 89%, which is what is expected under autotrophic conditions. Three bacterial groups are responsible for SNAD reactions: ammonium-oxidizing bacteria (AOB), anaerobic ammonium-oxidizing bacteria (AnAOB) and heterotrophic bacteria (HB). Because HB will compete with AOB and AnAOB for oxygen and nitrite, respectively, the system should be operated in such way that a balance among the different bacterial populations is achieved. Here, the results reported in the literature are analyzed to define suitable characteristics of effluents for treatment and operational conditions to allow the SNAD process to be carried out with different types of technologies.

Contents

1. Introduction

To fulfill disposal requirements, conventional nitrification–denitrification (ND) processes are generally used to remove both organic compounds and nitrogen from municipal and industrial wastewaters. These processes are well known, and their efficiency and reliability are beyond doubt. However, these processes have a major drawback: the availability of sufficient organic matter to carry out denitrification. Thus, ND processes can be applied only to effluents with chemical organic demand to nitrogen (COD/N) ratios higher than 5 [40]. The addition of an external organic carbon source required for COD/N ratios lower than 5 results in a consequent increase in operational costs [13]. Furthermore, application of the ND processes suggests that a portion of the organic matter present in the effluent is wasted by the aerobic route and cannot be used to produce biogas via anaerobic digestion [54]. To overcome this drawback, the application of partial nitrification-Anammox processes, instead of conventional ND

---

* Corresponding author at: Chemical Engineering Department, University of Concepción, Casilla 160-C, Concepción, Chile.

E-mail address: mroeckel@udec.cl (M.D. Roeckel).

http://dx.doi.org/10.1016/j.seppur.2016.04.012
1383-5866/

processes, is being promoted because these processes occur under autotrophic conditions, and organic matter and nitrogen can therefore be removed in separate processes [46,56].

Partial nitrification-Anammox processes involve two reactions in series. The first reaction (Eq. (1), [5] consists of aerobic oxidation of ammonium to nitrite by ammonia-oxidizing bacteria (AOB), and the second (Eq. (2), [39] is anoxic oxidation of ammonium to nitrogen gas using nitrite as an electron acceptor by the action of anaerobic ammonium-oxidizing bacteria (AnAOB).

$$NH_4^+ + 1.5O_2 \rightarrow NO_2^- + H_2O + 2H^+ \tag{1}$$

$$\begin{aligned} &NH_4^+ + 1.3NO_2^- + 0.066HCO_3^+ + 0.03H^+ \\ &\quad \rightarrow 1.02N_2 + 0.26NO_3^- + 0.066CH_2O_{0.5}N_{0.15} + 2H_2O \end{aligned} \tag{2}$$

To couple these reactions, approximately half of the ammonium must be oxidized to nitrite by partial nitrification; thus, an appropriate substrate for AnAOB is obtained, as shown in Eq. (3) [30].

$$NH_4^+ + 0.85O_2 \rightarrow 0.11NO_3^- + 0.445N_2 + 0.14H^+ + 1.43H_2O \tag{3}$$

Autotrophic nitrogen removal can be carried out in two-reactor systems, one for partial nitrification (PN) and a second for Anammox, or by coupling the reactions in a single reactor operated under controlled dissolved oxygen (DO) levels. The latter configuration is the one applied most frequently at an industrial scale because it involves less complex control systems, lower investment costs, reduced risk of AnAOB inhibition by nitrite and lower N_2O emissions [50]. In the past decade, a variety of technologies have been developed for performing PN-Anammox processes in a single-stage system, which can be divided into three types depending on the aggregation state of the biomass: suspended sludge, granular biomass and biofilm technologies.

Suspended sludge technologies utilize a mixed sludge containing flocculent bacteria (ammonia-oxidizing bacteria) and granular Anammox biomass. This technology includes a hydrocyclone to separate the granular Anammox bacteria from the flocculent biomass. The Anammox granules are returned to the reactor, whereas the flocs are separated and purged to avoid the proliferation of nitrite-oxidizing bacteria and thereby maintain a stable system operation [55]. When granular biomass or biofilms are used, AOB can grow in the outer part of the granule/biofilm and produce nitrite and consume oxygen, generating anoxic conditions in the inner part of the granule/biofilm. In this anoxic zone, ammonium and nitrite (produced during partial nitrification) must be present to allow the growth of Anammox bacteria [7,47].

Most of the industrial-scale applications of partial nitrification–Anammox (PN-Anammox) processes are installed in Europe, though there is growing interest in both China and North America [3]. Currently, there are more than 100 plants in operation worldwide, and the most used technology is that based on suspended biomass (approximately 40% of the plants), followed by granular systems and biofilms [26]. Most of the industrial-scale facilities treat the supernatant of anaerobic sludge digesters with inlet ammonium concentrations of 500–1500 mg NH_4^+-N/L and COD/NH_4^+-N ratios of 0.5–1.5 [26]. The high hydraulic residence time (HRT) applied to anaerobic sludge digesters (20–30 d) guarantees almost no remaining biodegradable COD in the effluent, which could interfere with the autotrophic process.

Nevertheless, the HRTs applied in cases of anaerobic digester treatment of industrial effluents are shorter than those used for anaerobic sludge digesters; therefore, the presence of relatively high amounts of biodegradable COD in the effluents cannot be overlooked [6,30,34,36,48]. A similar situation can occur during the application of PN-Anammox processes to the mainstream of municipal wastewater treatment plants (WWTPs) because organic matter is previously removed by an aerobic stage operated at a low solids retention time (SRT); thus, remaining biodegradable COD of approximately 20% of the inlet COD is commonly found due to the low SRT [14,29]. For this reason, the effect of organic matter on the partial nitrification and Anammox process has been gaining attention over the past few years [21,25,32].

2. Effect of organic matter on the partial nitrification and Anammox processes

Because PN-Anammox systems were initially implemented in a two-stage configuration, the first works investigating the effect of organic matter focused on the Anammox process [12,2]. Most research has shown that low organic levels do not significantly affect ammonia or nitrite removal but improve total nitrogen removal by denitrifiers [37]. In fact, the nitrate generated by Anammox bacteria can be reduced to nitrite in the presence of organic matter by heterotrophic denitrifying bacteria and can then be removed via the Anammox process [51]. However, if the inlet $COD_{biodegradable}/NO_2$-N ratio is greater than 1.9–3.1, Anammox bacteria are unable to compete with heterotrophic denitrifiers for both space and the electron acceptor (nitrite), with failed reactor performance [37,41,42]. Nevertheless, the coexistence of Anammox biomass with heterotrophic denitrifying biomass in the presence of organic matter has been reported [4]. This fact could be related to the capacity of Anammox bacteria to oxidize organic matter to CO_2 using nitrate and/or nitrite as the electron acceptor [16].

With regard to the effect of organic matter on partial nitrification, Mosquera-Corral et al. [36] found that an inlet $COD_{biodegradable}/NH_4^+$-N higher than 0.8 caused the washout of AOB from a chemostat operated at an SRT of 1 day and 30 °C due to competition between the heterotrophs and AOB. This negative effect of organic matter on suspended nitrifying biomass can be overcome by increasing the SRT [17]. Such findings would indicate that inlet $COD_{biodegradable}/NH_4^+$-N should be controlled to avoid AOB loss when the reactor is operated at SRT values close to the minimum SRT for AOB (0.5 d at 30 °C).

In the case of granular or biofilm nitrifying systems operated without devices to retain flocculent biomass, competition between heterotrophs and AOB can be avoided by HRT manipulation. If the HRT is larger than the reciprocal maximum specific growth rate of the heterotrophic biomass, the bacteria grow in suspension and do not form biofilms over the nitrifying biomass, which limits oxygen availability for ammonia oxidation [44]. If the HRT applied is shorter than the reciprocal maximum specific growth rate of the heterotrophic biomass, a heterotrophic layer will be formed on the nitrifying granular biomass or on the nitrifying biofilm, and its effect on the ammonia oxidation efficiency will depend not only on the $COD_{biodegradable}/NH_4^+$-N but also on the surface specific substrate load [43,44].

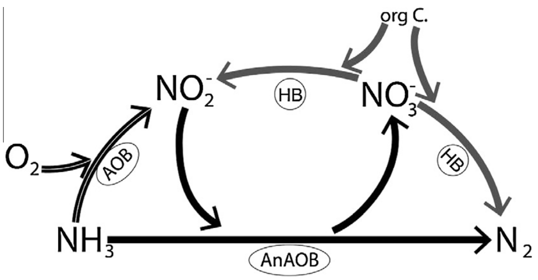

Fig. 1. SNAD process reactions. Different arrows indicate the different reactions mediated by AOB, AnAOB and HB, respectively.

Table 1
Operational conditions of different SNAD systems reported in the literature.

#	Reference	Reactor type/operation mode	Biomass/T° (°C)	Substrate	Inlet COD/N ratio	TRH (d)	TN and COD loading rates (kg/m^3·d)	Aeration regime	DO (mg/L)	Removal efficiency (%TN/ %COD)	Inlet (COD_{biod}/N)[*] ratio
1	Keluskar et al. [24]	Tubular up-flow reactor (1 L)/continuous	Suspended/30 °C	Effluent of a fertilizer industry. In mg/L: 700–800 TN, 46.6 COD	0.066	2.31	TN: 0.3-0.35 COD: 0.02	Continuous	2.9-3.9 ox. zone 0.1–0.4 anox. zone	98.9/46.6	0.03
2	Wang et al. [53]	Aeration tank (2.5 L)/SFBR	Suspended/35 °C	Landfill leachate. In mg/L: 295–700 TN, 250–500 COD	0.85 0.85 0.55 0.71	2.5	TN/COD: 0.118/0.1 0.24/0.2 0.26/0.15 0.28/0.2	Continuous	0.1	82/21 94/34 83/31 82/45	0.18 0.29 0.17 0.32
3	Lan et al. [27]	Aeration tank (18 L) SFBR	Suspended/35 °C	Synthetic wastewater. In mg/L: 200 TN, 100 COD	0.5	9/ 4.5/3	TN: 0.02–0.067 COD: 0.01–0.03	Continuous	0.5–1	93–96/72–87	0.36–0.44
4	Joss et al. [22]/ Langone et al. [28]	Aeration tank (1400 m^3)/ SFBR	Suspended/30 °C	Digested supernatant of a municipal WWTP. In mg/L: 650 TN, 300 COD	0.5 approx.	0.78	TN: 0.83 COD: 0.38	Continuous	0.1–1	94.6/37	0.17
5	Daverey et al. [9]	Aeration tank (5 L)/SBR – SFBR	Granules / 15–30 °C (without control)	Anaerobic digester liquor of swine wastewater. In mg/L: 662 ± 190 TN, 387 ± 145 COD	0.42 2.56	5/2.5	TN: 0.045–0.175 COD: 0.026–0.236	Continuous	<0.5	Batch: 52/57 Fed batch: 80/76	0.3 0.9
6	Daverey et al. [8]	Aeration tank (2.5 L)/SFBR	Granules/25 °C	Opto-electronic wastewater. In mg/L: 572 ± 6.6 TN, 100 ± 28 COD	0.17	2.5	TN: 0.23 COD: 0.04	Continuous	0.1	83–93/79	0.13
7	Winkler et al. [57]	Bubble column (2.9 L)/SBR	Granules/18 °C	Synthetic wastewater. In mg/L: 200 TN, 100 COD	0.5	0.32	TN: 0.62 COD: 0.31	Cycles ox/anox: 172 min/60 min	1.5 (oxic phase)	90/100	0.5
8	Wang et al. [52]	Aeration tank (384 m^3)/continuous	Granules/30–33 °C	Landfill leachate. In mg/L: 634 TN, 554 COD	0.87	1.26	TN: 0.5 COD: 0.44	Continuous	0.3	76/28	0.24
9	Daverey et al. [10]	Biofilm reactor (2.5 L)/SBR	Biofilm / 25 °C	Synthetic wastewater. In mg/L: 600 TN, 300 COD	0.5	1.67	TN: 0.36 COD: 0.18	Continuous	0.1	88/90	0.44
10	Zhao et al. [60]	Biofilm reactor (5 L)/SBR	Biofilm / 32 °C	Synthetic wastewater. In mg/L: 428 TN, 150 COD	0.35	0.78	TN: 0.55 COD: 0.19	Continuous	1–1.5	88.8/∼50	0.175
11	Liang et al. [30]	Packing up-flow biofilter (2.65 L) / continuous	Biofilm/ 25 °C	Synthetic wastewater. In mg/L: 200 TN, 40 COD	0.2	0.025	TN: 8 COD: 1.6	Continuous (4.5 L air/min)	Not reported	76/81	0.162
12	Zhang et al. [58]	Packing biofilter (6.5 L) / SBR	Biofilm/30 °C	Swine digester liquor. In mg/L: 437 ± 4 COD, 472 ± 6 TN	0.81 0.65 1.24	1	TN/COD: 0.20/0.16 0.25/0.16 0.20/0.25	Cycles ox/anox: 3 h/1 h	1.5–2 ox. cycle 0.2–0.5 anox. cycle	47/39 62/39 29/56	0.32 0.25 0.69
13	Chen et al. [6]	NRBC[b]/continuous	Biofilm/35 °C	Synthetic wastewater. In mg/L: 200 TN, 150 and 100 COD	0.75 0.5	0.29	TN: 0.69 COD: 0.34	Continuous	0.4–0.6	52/68 70/94	0.47–0.71

(a) non-woven rotating biological contactor.
SBR: Sequencing Batch Reactor; SFBR: Sequencing Fed Batch Reactor; TN: Total Nitrogen.
[*] The COD_{biod}/N ratio was calculated based on the COD effectively degraded inside the reactor and the inlet N.

When both PN and Anammox processes are carried out in one stage and in the presence of organic matter, destabilization of the nitrogen removal process could be expected due to the development of heterotrophic bacteria, which can displace AOB and AnAOB by competing for oxygen and nitrite, respectively, due to higher growth rates [58,30]. Nevertheless, if suitable operational conditions and inlet $COD_{biodegradable}/N$ ratios are provided, a balance among AOB, AnAOB and HB activities can be achieved, thereby maintaining high nitrogen removal efficiency. According to Eq. (3), coupled reactions of partial nitrification and Anammox are capable of removing up to 89% of ammonium, leaving the remaining 11% of nitrogen in the form of nitrate. In the presence of organic matter, the remaining nitrate can be used by HB as an electron acceptor for organic carbon oxidation, which would allow the theoretical removal of 100% of the nitrogen by the combined action of these three bacterial groups (Eq. (4)). This new process has been called the simultaneous nitrification, Anammox and denitrification (SNAD) process (Fig. 1).

Since its emergence in 2009 [6], an abrupt increase in the number of published articles on the SNAD process has occurred compared with other N removal processes [59]. In the next sections, the fundamental aspects of operational strategies for maintaining a stable operation and reactor performance improvement are discussed.

$$NO_3^- + 1.08CH_3OH + 0.24H_2CO_3 \rightarrow 0.056C_5H_7O_2N + 0.47N_2 + 1.68H_2O + HCO_3^- \quad (4)$$

3. Analysis of SNAD process performance

Much information about the performance of the SNAD process can be found in the literature (Table 1). Although this process has been applied to different types of effluents using different types of technologies and operational conditions, its operational limits for maintaining high N removal efficiency have not yet been defined. For this reason, the main aspects that can affect competition between HB and AOB/AnAOB, such as the COD/N influent ratio, solid retention time (SRT) and aeration regime, will be analyzed in the ensuing sections. The microbial characteristics of SNAD systems are also described.

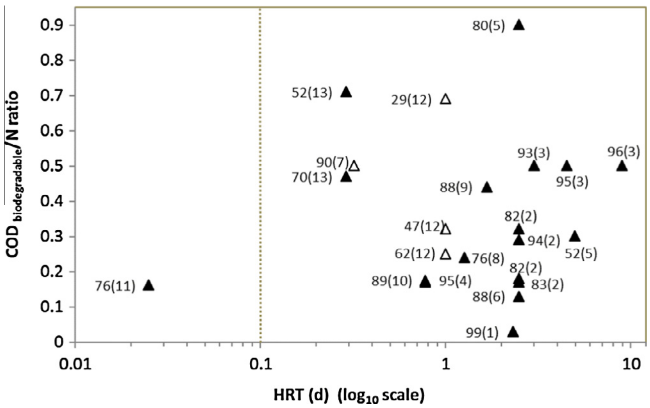

Fig. 2. Plot of the solids retention time (SRT) and $COD_{biodegradable}/N$ ratios utilized by different authors for the SNAD process. Triangles are accompanied by N removal efficiency (%) obtained in each experiment, and the number in parenthesis indicates the reference: (1) [24]; (2) Wang et al. [53]; (3) Lan et al. [27]; (4) Joss et al. [22]/ Langone et al. [28]; (5) Daverey et al. [9]; (6) Daverey et al. [8]; (7) Winkler et al. [57]; (8) Wang et al. [52]; (9) Daverey et al. [10]; (10) Zhao et al. [60]; (11) Liang et al. [30]; (12) Zhang et al. [58]; and (13) Chen et al. [6]. Filled triangles: continuous aeration mode; unfilled triangles: intermittent aeration mode.

3.1. COD/N influent ratio

To date, effluents from anaerobic digesters treating swine wastewater [9,58] and sludge [28] from the fertilizer industry [24], opto-electronic wastewater [8] and landfill leachate [52,53] have been tested in SNAD reactors.

In general, the inlet COD/N ratio reported in the literature takes into account the total COD; however, only the biodegradable fraction of organic matter should be considered because it is the available substrate for heterotrophic growth. For this reason, for the comparison of different works, the inlet COD/N ratio was calculated based on the organic matter degraded inside the reactor ($COD_{biodegradable}/N$). As shown in Fig. 2, almost all SNAD studies have be conducted with effluents having a $COD_{biodegradable}/N$ ratio lower than 0.7; therefore, no negative effects on the Anammox biomass or the AOB are expected [36,42]. These low applied $COD_{biodegradable}/N$ ratios could explain the good performance of most of the SNAD experiments to date.

3.2. Applied solids retention time

Despite the importance of the SRT on the performance of biological systems, few works have provided the SRT values applied to SNAD reactors [22,52]. Nonetheless, authors do provide the HRT values applied to their systems. For suspended biomass systems, the HRT ranges between 2 and 5 d, whereas these values for granular and biofilm systems range between 1 and 3 d and 0.025–1 d, respectively (Fig. 2).

As mentioned previously, the SRT of the flocculent biomass in granular and biofilm systems is crucial to avoid heterotrophic growth over the nitrifying biomass which limits oxygen availability for ammonia oxidation. Thus, to separate autotrophic and heterotrophic biomasses spatially in granular and biofilm systems, the SRT imposed on the flocculent biomass should be larger than the reciprocal maximum specific growth rate of the heterotrophic biomass (approximately 0.1 days at 30 °C [45]). Because SRT is equal to or larger than HRT, the development of a layer of heterotrophic biomass over biofilms could be expected only in the work of Liang et al. [30] (point 11 of Fig. 2), in which a biofilm system was operated at an HRT of 0.025 d. Nevertheless, this system was able to operate with a nitrogen removal efficiency of 76%, most likely due to the low $COD_{biodegradable}/N$ ratio of the influent (0.16).

3.3. Dissolved oxygen level and aeration strategy

Under autotrophic conditions, the theoretical nitrogen removal efficiency of the PN-Anammox process is 89%, though the efficiency obtained at an industrial scale is approximately 80–85% [26]. Therefore, if the SNAD process is carried out properly, efficiencies higher than the latter values are expected. Most SNAD processes have been performed under continuous aeration at DO levels between 0.1 O_2/L and 0.5 mg O_2/L, which are suitable for simultaneous nitrification and denitrification [18]. The nitrogen removal efficiency reported for these systems is 88 ± 8% (calculated from the data shown in Table 1). Therefore, the promotion of simultaneous nitrification and denitrification under microaerobic conditions enhances the efficiency of the system. However,

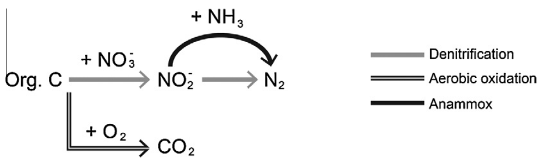

Fig. 3. Organic carbon oxidation pathways.

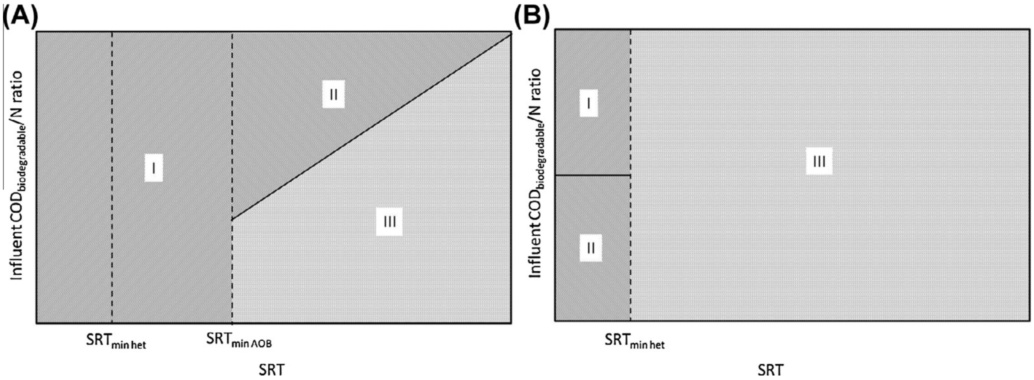

Fig. 4. Different operational zones depending on the influent COD$_{biodegradable}$/N ratio and on the solid retention time (SRT) applied to flocculent biomass (heterotrophic biomass in the case of granular and biofilm systems and heterotrophic and ammonia-oxidizing biomass in the case of suspended biomass systems). (A) Suspended biomass systems: zone I – washout of AOB; zone II – proliferation of heterotrophic biomass; and zone III – stable operation. (B) Granular biomass and biofilm systems: zone I – formation of heterotrophic biofilm causing oxygen limitation; zone II – formation of heterotrophic biofilm not causing oxygen limitation; and zone III – stable operation.

continuous aeration also causes a portion of the organic matter to be oxidized by oxygen instead of nitrate, which increases the operational costs. In this sense, the alternation of oxic and anoxic periods to promote the use of the organic matter present for denitrification would reduce aeration costs and increase the nitrogen removal efficiency [58,57]. Thus, heterotrophic denitrification occurs during the anoxic phase, consuming most of the organic carbon and reducing the nitrate produced by Anammox; this in turn allows PN-Anammox processes to occur practically in the absence of organic carbon in the following oxic phase, during which ammonia is converted to N_2 by the combined action of AOB and AnAOB (Fig. 3). Depending on the availability of a carbon source during the anoxic period, nitrate can be completely reduced to N_2 or partially reduced to nitrite, which is consumed by the Anammox bacteria (Fig. 3). During the oxic phase, denitrification is inhibited and organic carbon is preferentially consumed by the aerobic route, a time when oxygen levels are higher than 0.5 mg O_2/L (Fig. 3).

Winkler et al. [57] used a granular SBR reactor initially fed with wastewater and operated under anoxic conditions to remove nitrate and organic matter via heterotrophic denitrification; air was then supplied (DO: 1.5 mg O_2/L) to promote nitrogen removal by autotrophic bacteria. This system achieved a nitrogen removal efficiency of 90%, which was most likely limited by the volumetric exchange ratio applied (50%). Nevertheless, Zhang et al. [58] proposed a cycle in which wastewater was initially fed, and the system was then operated under aerobic conditions (1.5–2 mg O_2/L), followed by an anoxic period. In this case, the nitrogen removal efficiency was 29–62%. This would indicate that better results are achieved when organic matter is mainly consumed under anoxic conditions.

If alternating oxic/anoxic conditions are applied in suspended biomass systems, the minimum SRT applied to the flocculent biomass should be increased with respect to those systems continuously aerated, as AOB can grow only during oxic periods. In the case of granular or biofilm systems, the minimum SRT applied to heterotrophic biomass will not be affected when compared to continuously aerated systems because this biomass can grow under both oxic and anoxic conditions.

3.4. Microbial community distribution

The coexistence of AOB, AnAOB and HB communities has been proven in SNAD systems with high N removal efficiencies [1,6,28]. In addition, stratified distribution of different bacterial groups has been observed, with AnAOB and denitrifiers being more abundant in anoxic zones [6] and AOB predominantly in oxic zones [6,24]. However, the proportion of each type of bacterium has rarely been described. Liang et al. [30] detected a reduction in the relative abundance of AOB and AnAOB from 35% and 40% to 30% and 33%, respectively, with COD addition in a CANON biofilm system, most likely due to the development of HB, such as denitrifiers. Keluskar et al. [24] detected different abundances of AOB and denitrifiers in a tubular up-flow reactor with suspended biomass, with AOB being more abundant in the oxic zone (61% versus 6% in the anoxic zone) and denitrifiers more abundant in the anoxic zone (22% versus 10% in the oxic zone). The dominance of AnAOB in the anoxic zone was also suggested due to the detection of uncultured Planctomycetes in the biomass amplified. It appears that the functional proportion and distribution of different bacterial groups depends on the characteristics of the system, though further studies on microbial dynamics must be performed.

4. Conclusions

Most of the SNAD systems reported in the literature have achieved high nitrogen removal efficiencies. These systems have applied different technologies, were fed with different types of effluents and were operated under different operational conditions. However, the operational limits that guarantee the stability of the process are not well defined to date. In this sense, similar operational considerations to those used for maintaining the stability of the systems under autotrophic conditions must be taken into account [35], in addition to other considerations related to establishing a balance between heterotrophic and autotrophic populations:

- SNAD systems based on suspended biomass should be operated in such a way that the SRT applied to the flocculent biomass (heterotrophic and ammonia-oxidizing biomass) is sufficient to allow appropriate ammonia oxidation activity (Zone III, Fig. 4A) and to avoid the washout of AOB (Zone I, Fig. 4A). An effluent with a relatively high COD$_{biodegradable}$/N ratio could cause the proliferation of heterotrophic biomass, which should be purged from the system. This would suggest a reduction in the SRT of the flocculent biomass and therefore a decrease in the ammonia oxidation capacity. Moreover, if the SRT applied to the flocculent biomass is close to the minimum SRT required by AOB, the development of heterotrophic biomass will have a more negative effect on the ammonia oxidation capacity (Zone II, Fig. 4A).
- SNAD systems based on granular biomass or biofilms should be operated in such a way that heterotrophic biomass is allowed to grow in suspension by applying an SRT higher than 0.1 d for

systems operated at 30 °C (Zone III, Fig. 4B). In this way, the risk of process stability loss due to oxygen limitations during ammonia oxidation by a heterotrophic biofilm is minimized. This risk will also depend on the $COD_{biodegradable}/N$ ratio of the influent and the applied specific surface-loading rate. Therefore, operation at an HRT smaller than 0.1 d and a relatively high $COD_{biodegradable}/N$ ratio is not advisable for this type of system (Zone I, Fig. 4B), whereas an HRT smaller than 0.1 d and a relatively low $COD_{biodegradable}/N$ ratio will promote the growth of heterotrophic biomass on the granular biomass or biofilm, with oxygen still being available for AOB (Zone II, Fig. 4B).

To improve the nitrogen removal efficiency of the system, alternating oxic/anoxic periods could be implemented to promote organic carbon consumption via denitrification. To achieve this, the feeding strategy must promote the presence of organic carbon during anoxic periods.

5. Prospects

As mentioned above, most of the applications of the SNAD process have been focused on treating effluents from industrial anaerobic digesters containing high ammonia concentrations at a temperature of approximately 30 °C. However, due to the recent interest in improving the energy efficiency of WWTPs by means of the application of Anammox-based processes in the mainstream, future efforts will be addressed at studying the stability of the SNAD process operated at ammonia concentrations of approximately 50 mg N/L and ambient temperatures [57,33,11]. COD can be mainly removed from mainstream by an aerobic reactor operated at a low SRT to save oxygen and to maximize methane production via sludge digesters; the main effluent with an expected COD/N ratio of approximately 2 [29,31,33] would then be treated via the SNAD process. Because Anammox bacteria have the slowest growth rate among the microorganisms involved in the SNAD process, they could be expected to be the bottleneck of this process. Nevertheless, several works have demonstrated that Anammox bacteria can maintain a stable activity even at temperatures between 10 and 20 °C [19,15,20,49] and in the presence of organic matter [29,31]. Moreover, the ability of AnAOB to reduce nitrate to nitrite using short-chain fatty acids [16,23] could contribute to improving the effluent quality in terms of nitrogen compounds [38]. Recent studies report that the actual limitation of SNAD application to the mainstream of WWTPs appears to be the proliferation of nitrite-oxidizing bacteria, which contribute to an excessive presence of nitrate in the effluent. New strategies have been proposed for partial nitrification to suppress nitrite oxidizing bacteria, as alternating aerobic/anoxic conditions. However, the full scale application of this strategy implies a precise design of operation conditions as oxic/anoxic cycles, different solids retention times for each type of biomasses and a strict control of the dissolved oxygen and ammonia concentrations [35].

References

[1] R. Abbassi, A.K. Yadav, S. Huang, P.R. Jaffé, Laboratory study of nitrification, denitrification and anammox processes in membrane bioreactors considering periodic aeration, J. Environ. Manage. 142 (2014) 53–59.

[2] Y. Ahn, I. Hwang, K. Min, ANAMMOX and partial denitritation in anaerobic nitrogen removal from piggery waste, Water Sci. Technol. 49 (5–6) (2004) 145–153.

[3] M. Ali, S. Okabe, Anammox-based technologies for nitrogen removal: advances in process start-up and remaining issues, Chemosphere 141 (2015) 144–153.

[4] G. Anjali, P.C. Sabumon, Unprecedented development of Anammox in presence of organic carbon using seed biomass from a tannery Common Effluent Treatment Plant (CETP), Bioresour. Technol. 153 (2014) 30–38.

[5] D.J. Arp, L.Y. Stein, Metabolism of inorganic N compounds by ammonia-oxidizing bacteria, Crit. Rev. Biochem. Mol. Biol. 38 (6) (2003) 471–495.

[6] H. Chen, S. Liu, F. Yang, Y. Xue, T. Wang, The development of simultaneous partial nitrification, ANAMMOX and denitrification (SNAD) process in a single reactor for nitrogen removal, Bioresour. Technol. 100 (4) (2009) 1548–1554.

[7] M. Christensson, S. Ekström, A.A. Chan, E. Le Vaillant, R. Lemaire, Experience from start-ups of the first ANITA Mox plants, Water Sci. Technol. 67 (12) (2013) 2677–2684.

[8] A. Daverey, S.H. Su, Y.T. Huang, J.G. Lin, Nitrogen removal from opto-electronic wastewater using the simultaneous partial nitrification, anaerobic ammonium oxidation and denitrification (SNAD) process in sequencing batch reactor, Bioresour. Technol. 113 (2012) 225–231.

[9] A. Daverey, N.T. Hung, K. Dutta, J.G. Lin, Ambient temperature SNAD process treating anaerobic digester liquor of swine wastewater, Bioresour. Technol. 141 (2013) 191–198.

[10] A. Daverey, Y.C. Chen, K. Dutta, Y.T. Huang, J.G. Lin, Start-up of simultaneous partial nitrification, anammox and denitrification (SNAD) process in sequencing batch biofilm reactor using novel biomass carriers, Bioresour. Technol. 190 (2015) 480–486.

[11] H. De Clippeleir, S.E. Vlaeminck, F. De Wilde, K. Daeninck, M. Mosquera, P. Boeckx, N. Boon, One-stage partial nitritation/anammox at 15 °C on pretreated sewage: feasibility demonstration at lab-scale, Appl. Microbiol. Biotechnol. 97 (23) (2013) 10199–10210.

[12] X. Dong, E.W. Tollner, Evaluation of Anammox and denitrification during anaerobic digestion of poultry manure, Bioresour. Technol. 86 (2) (2003) 139–145.

[13] C. Fux, H. Siegrist, Nitrogen removal from sludge digester liquids by nitrification/denitrification or partial nitritation/anammox: environmental and economical considerations, Water Sci. Technol. 50 (10) (2004) 19–26.

[14] H. Ge, D.J. Batstone, J. Keller, Operating aerobic wastewater treatment at very short sludge ages enables treatment and energy recovery through anaerobic sludge digestion, Water Res. 47 (17) (2013) 6546–6557.

[15] E.M. Gilbert, S. Agrawal, S.M. Karst, H. Horn, P.H. Nielsen, S. Lackner, Low temperature partial nitritation/anammox in a moving bed biofilm reactor treating low strength wastewater, Environ. Sci. Technol. 48 (15) (2014) 8784–8792.

[16] D. Güven, A. Dapena, B. Kartal, M.C. Schmid, B. Maas, K. van de Pas-Schoonen, S. Sosen, R. Mendez, H. Op den Camp, M. Jetten, M. Strous, I. Schmidt, Propionate oxidation by and methanol inhibition of anaerobic ammonium-oxidizing bacteria, Appl. Environ. Microbiol. 71 (2) (2005) 1066–1071.

[17] K. Hanaki, C. Wantawin, S. Ohgaki, Effects of the activity of heterotrophs on nitrification in a suspended-growth reactor, Water Res. 24 (3) (1990) 289–296.

[18] S.M. Hocaoglu, G. Insel, E.U. Cokgor, D. Orhon, Effect of low dissolved oxygen on simultaneous nitrification and denitrification in a membrane bioreactor treating black water, Bioresour. Technol. 102 (6) (2011) 4333–4340.

[19] Z. Hu, T. Lotti, M. de Kreuk, R. Kleerebezem, M. van Loosdrecht, J. Kruit, B. Kartal, Nitrogen removal by a nitritation-anammox bioreactor at low temperature, Appl. Environ. Microbiol. 79 (8) (2013) 2807–2812.

[20] K. Isaka, Y. Date, Y. Kimura, T. Sumino, S. Tsuneda, Nitrogen removal performance using anaerobic ammonium oxidation at low temperatures, FEMS Microbiol. Lett. 282 (1) (2008) 32–38.

[21] R.C. Jin, G.F. Yang, J.J. Yu, P. Zheng, The inhibition of the Anammox process: a review, Chem. Eng. J. 197 (2012) 67–79.

[22] A. Joss, D. Salzgeber, J. Eugster, R. König, K. Rottermann, S. Burger, P. Fabijan, S. Leumann, J. Mohn, H. Siegrist, Full-scale nitrogen removal from digester liquid with partial nitritation and anammox in one SBR, Environ. Sci. Technol. 43 (14) (2009) 5301–5306.

[23] B. Kartal, L. Van Niftrik, J. Rattray, J.L. Van De Vossenberg, M.C. Schmid, J.S. Damsté, M. Strous, Candidatus 'Brocadia fulgida': an autofluorescent anaerobic ammonium oxidizing bacterium, FEMS Microbiol. Ecol. 63 (1) (2008) 46–55.

[24] R. Keluskar, A. Nerurkar, A. Desai, Development of a simultaneous partial nitrification, anaerobic ammonia oxidation and denitrification (SNAD) bench scale process for removal of ammonia from effluent of a fertilizer industry, Bioresour. Technol. 130 (2013) 390–397.

[25] M. Kumar, J.G. Lin, Co-existence of anammox and denitrification for simultaneous nitrogen and carbon removal—strategies and issues, J. Hazard. Mater. 178 (1) (2010) 1–9.

[26] S. Lackner, E.M. Gilbert, S.E. Vlaeminck, A. Joss, H. Horn, M.C. van Loosdrecht, Full-scale partial nitritation/anammox experiences–an application survey, Water Res. 55 (2014) 292–303.

[27] C.J. Lan, M. Kumar, C.C. Wang, J.G. Lin, Development of simultaneous partial nitrification, anammox and denitrification (SNAD) process in a sequential batch reactor, Bioresour. Technol. 102 (9) (2011) 5514–5519.

[28] M. Langone, J. Yan, S.C. Haaijer, H.J.O. den Camp, M.S. Jetten, G. Andreottola, Coexistence of nitrifying, anammox and denitrifying bacteria in a sequencing batch reactor, Front. Microbiol. 5 (2014).

[29] M. Laureni, D.G. Weissbrodt, I. Szivák, O. Robin, J.L. Nielsen, E. Morgenroth, A. Joss, Activity and growth of anammox biomass on aerobically pre-treated municipal wastewater, Water Res. 80 (2015) 325–336.

[30] Y. Liang, D. Li, X. Zhang, H. Zeng, Z. Yang, J. Zhang, Microbial characteristics and nitrogen removal of simultaneous partial nitrification, anammox and denitrification (SNAD) process treating low C/N ratio sewage, Bioresour. Technol. 169 (2014) 103–109.

[31] T. Lotti, R. Kleerebezem, C. van Erp Taalman Kip, T.L. Hendrickx, J. Kruit, M. Hoekstra, M.C. van Loosdrecht, Anammox growth on pretreated municipal wastewater, Environ. Sci. Technol. (14) (2014) 7874–7880.

[32] A. Magrí, F. Béline, P. Dabert, Feasibility and interest of the anammox process as treatment alternative for anaerobic digester supernatants in manure processing–an overview, J. Environ. Manage. 131 (2013) 170–184.

[33] A. Malovanyy, J. Trela, E. Plaza, Mainstream wastewater treatment in integrated fixed film activated sludge (IFAS) reactor by partial nitritation/anammox process, Bioresour. Technol. 198 (2015) 478–487.

[34] B. Molinuevo, M.C. García, D. Karakashev, I. Angelidaki, Anammox for ammonia removal from pig manure effluents: effect of organic matter content on process performance, Bioresour. Technol. 100 (7) (2009) 2171–2175.

[35] N. Morales, Á.V. del Río, J.R. Vázquez-Padín, R. Méndez, A. Mosquera-Corral, J.L. Campos, Integration of the Anammox process to the rejection water and main stream lines of WWTPs, Chemosphere 140 (2015) 99–105.

[36] A. Mosquera-Corral, F. Gonzalez, J.L. Campos, R. Méndez, Partial nitrification in a SHARON reactor in the presence of salts and organic carbon compounds, Process Biochem. 40 (9) (2005) 3109–3118.

[37] S.Q. Ni, J.Y. Ni, D.L. Hu, S. Sung, Effect of organic matter on the performance of granular anammox process, Bioresour. Technol. (2012) 701–705.

[38] P. Regmi, B. Holgate, M.W. Miller, H. Park, K. Chandran, B. Wett, C.B. Bott, Nitrogen polishing in a fully anoxic anammox MBBR treating mainstream nitritation–denitritation effluent, Biotechnol. Bioeng. (2015).

[39] M. Strous, J.J. Heijnen, J.G. Kuenen, M.S.M. Jetten, The sequencing batch reactor as a powerful tool for the study of slowly growing anaerobic ammonium-oxidizing microorganisms, Appl. Microbiol. Biotechnol. 50 (5) (1998) 589–596.

[40] S.P. Sun, C.P.I. Nàcher, B. Merkey, Q. Zhou, S.Q. Xia, D.H. Yang, J.H.S. Sun, B.F. Smets, Effective biological nitrogen removal treatment processes for domestic wastewaters with low C/N ratios: a review, Environ. Eng. Sci. 27 (2) (2010) 111–126.

[41] C.J. Tang, P. Zheng, C.H. Wang, Q. Mahmood, Suppression of anaerobic ammonium oxidizers under high organic content in high-rate Anammox UASB reactor, Bioresour. Technol. 101 (6) (2010) 1762–1768.

[42] C.J. Tang, P. Zheng, L.Y. Chai, X.B. Min, Thermodynamic and kinetic investigation of anaerobic bioprocesses on ANAMMOX under high organic conditions, Chem. Eng. J. 230 (2013) 149–157.

[43] L. Tijhuis, E. Rekswinkel, M.C.M. Van Loosdrecht, J.J. Heijnen, Dynamics of population and biofilm structure in the biofilm airlift suspension reactor for carbon and nitrogen removal, Water Sci. Technol. 29 (1994) 377–384.

[44] W.A.J. Van Benthum, M.D.M. Van Loosdrecht, J.J. Heijnen, Control of heterotrophic layer formation on nitrifying biofilms in a biofilm airlift suspension reactor, Biotechnol. Bioeng. 53 (4) (1997) 397–405.

[45] W.V. Van Benthum, J.M. Garrido, J.P.M. Mathijssen, J. Sunde, M.V. Loosdrecht, J. J. Heijnen, Nitrogen removal in intermittently aerated biofilm airlift reactor, J. Environ. Eng. 124 (3) (1998) 239–248.

[46] M.C.M. Van Loosdrecht, X. Hao, M.S.M. Jetten, W. Abma, Use of Anammox in urban wastewater treatment, Water Sci. Technol.: Water Supply 4 (1) (2004) 87–94.

[47] J. Vázquez-Padín, I. Fernádez, M. Figueroa, A. Mosquera-Corral, J.L. Campos, R. Méndez, Applications of Anammox based processes to treat anaerobic digester supernatant at room temperature, Bioresour. Technol. 100 (12) (2009) 2988–2994.

[48] J.R. Vázquez-Padın, M. Figueroa, I. Fernández, A. Mosquera-Corral, J.L. Campos, R. Mendez, Post-treatment of effluents from anaerobic digesters by the Anammox process, Water Sci. Technol. 60 (2009) 1135–1143.

[49] J.R. Vazquez-Padın, I. Fernández, N. Morales, J.L. Campos, A. Mosquera-Corral, R. Méndez, Autotrophic nitrogen removal at low temperature, Water Sci. Technol. 9 (63.6) (2011) 9.

[50] S.E. Vlaeminck, H. De Clippeleir, W. Verstraete, Microbial resource management of one-stage partial nitritation/anammox, Microb. Biotechnol. 5 (3) (2012) 433–448.

[51] M. Waki, T. Yasuda, Y. Fukumoto, K. Kuroda, K. Suzuki, Effect of electron donors on anammox coupling with nitrate reduction for removing nitrogen from nitrate and ammonium, Bioresour. Technol. 130 (2013) 592–598.

[52] C.C. Wang, P.H. Lee, M. Kumar, Y.T. Huang, S. Sung, J.G. Lin, Simultaneous partial nitrification, anaerobic ammonium oxidation and denitrification (SNAD) in a full-scale landfill-leachate treatment plant, J. Hazard. Mater. 175 (1) (2010) 622–628.

[53] C.C. Wang, M. Kumar, C.J. Lan, J.G. Lin, Landfill-leachate treatment by simultaneous partial nitrification, anammox and denitrification (SNAD) process, Desalin. Water Treat. 32 (1–3) (2011) 4–9.

[54] B. Wett, Development and implementation of a robust deammonification process, Water Sci. Technol. 56 (7) (2007) 81–88.

[55] B. Wett, K. Buchauer, C. Fimml, Energy self-sufficiency as a feasible concept for wastewater treatment systems, Asian Water (September) (2007) 22–25.

[56] B. Wett, A. Omari, S.M. Podmirseg, M. Han, O. Akintayo, M. Gómez-Brandón, M. O'Shaughnessy, Going for mainstream deammonification from bench-to full-scale for maximized resource efficiency, Water Sci. Technol. 68 (2013) 283–289.

[57] M.K. Winkler, R. Kleerebezem, M.C.M. Van Loosdrecht, Integration of anammox into the aerobic granular sludge process for main stream wastewater treatment at ambient temperatures, Water Res. 46 (1) (2012) 136–144.

[58] Z. Zhang, Y. Li, S. Chen, S. Wang, X. Bao, Simultaneous nitrogen and carbon removal from swine digester liquor by the Canon process and denitrification, Bioresour. Technol. 114 (2012) 84–89.

[59] Z. Zhang, S. Liu, Hot topics and application trends of the anammox biotechnology: a review by bibliometric analysis, SpringerPlus 3 (1) (2014) 1–8.

[60] J. Zhao, J. Zuo, J. Lin, P. Li, The performance of a combined nitritation–anammox reactor treating anaerobic digestion supernatant under various C/N ratios, J. Environ. Sci. 30 (2015) 207–214.