Desalination and Water Treatment
www.deswater.com

doi: 10.1080/19443994.2016.1235152

57 (2016) 27890–27898
December

Taylor & Francis
Taylor & Francis Group

A critical review of future trends and perspectives for the implementation of partial nitritation/anammox in the main line of municipal WWTPs

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Received 30 October 2015; Accepted 3 August 2016

ABSTRACT

The use of technologies based on partial nitritation (PN) and anaerobic ammonium oxidation (Anammox) is a cost-efficient and sustainable alternative for nitrogen removal from wastewaters with low COD/N ratios. These technologies allow savings in biodegradable organic matter consumption; oxygen requirements (about 40% lower compared to the conventional process of nitrification/denitrification); and sludge treatment costs. Despite the advantages of the PN/Anammox process, it also has some limitations which led to a full-scale application relatively restricted to some wastewaters, especially the reject water line of wastewater treatment plants (WWTPs). However, the PN/Anammox process also opens the interesting possibility of transforming most of the biodegradable organic matter arriving at the WWTP into biogas, because that biodegradable COD is not necessary anymore to denitrify. This can lead to an energetically self-sufficient WWTP, which means important cost savings. The main aspects arising from this application would be: the treatment of low-strength wastewater with high variability in concentrations and loads; the possible effects of biodegradable COD reaching the PN/Anammox system; the operation at low temperatures; the effective retention of microbial populations of interest; the use of one-step or two-step systems; and the effective control of the process during the start-up and the stable operation. Some of the proposed solutions which will be detailed along this paper are: the use of biofilm biomass (with or without carrier); novel aeration strategies and control systems; and different reactor configurations.

Keywords: Anammox; Biological nutrient removal; Energetic self-sufficiency; Municipal wastewater; Sustainability

1. Introduction

The use of technologies based on anaerobic ammonium oxidation (Anammox) is a cost-efficient and sustainable alternative for nitrogen removal from wastewaters with low COD/N ratios [1,2]. The Anammox bacteria are autotrophic, thus biodegradable organic matter is not necessary. In addition, since the substrates of Anammox sludge are nitrite and ammonium, the requirements of oxygen are about 40% lower compared to the conventional process (i.e. nitrification/denitrification) given that only about 50% of the ammonium needs to be oxidized to nitrite. The production of sludge decreases to a great extent

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compared to the conventional process due to the small biomass yield of the autotrophic Anammox organisms [3]. Taking this into account, a cost evaluation [4] gave an estimation of about 0.22 €/kg N for a partial nitritation (PN)/Anammox treatment, which agrees with the one reported by Van Dongen et al. [5], vs. about 0.66 €/kg N for a conventional nitrification/denitrification process. Furthermore, Siegrist et al. [6] reported about a 50% of energy savings with the implementation of the PN/Anammox process in a municipal wastewater treatment plant (WWTP), achieving an energy consumption of 0.021 kWh/(p.e. d).

Despite the advantages of the Anammox process, it also has some limitations [2], e.g. inhibition by substrates and exogenous compounds including biodegradable COD [7], optimum temperature in the mesophile range [8] and slow start-up [9]. Thus, its full-scale application is still relatively restricted to wastewaters from mesophilic anaerobic digesters, especially in the reject water line of WWTPs. In fact, Lackner et al. [10] reported that about 75% of the full-scale PN/Anammox reactors (as of 2014) were operated for side stream treatment of municipal wastewater. Furthermore, these authors [10] reported that the majority of the full-scale PN/Anammox systems, both at municipal or industrial WWTPs, are treating high-strength wastewaters with ammonium concentrations around 1 g N/L, which are far higher compared to the usual ammonium concentrations (20–75 mg NH_4^+-N/L [11]) in municipal wastewaters.

However, the Anammox process also leads to the interesting possibility of converting most of the biodegradable organic matter arriving at the plant into biogas (i.e. methanization), because that biodegradable COD is not necessary anymore to denitrify [12]. This can head to an energetically self-sufficient (or even energy-generating) WWTP, which means important cost savings [13,14]. This application was already proposed by Jetten et al. [15] as early as almost two decades ago. However, it has been barely implemented for the moment [16,17]. Therefore, the main focus of this work is reviewing the latest advances on the application of the PN/Anammox process in the main line and the existing limitations. This topic is already a hotspot of the wastewater treatment research because of its potential to improve the existing plants and the design of the new ones, lowering costs and environmental footprints [13–15]. Therefore, this literature review may help researchers and, eventually, plant operators to know the present state of the topic and its research trends. Specifically, the most important issues which will be discussed are: the management of high COD/N ratio (pretreatment); the treatment of low-strength wastewater; the operation at low temperatures; the effective retention of microbial populations of interest; and the use of one-step or two-step systems [12]. Some of the proposed solutions which will be detailed are: physicochemical or biological methods to concentrate the biodegradable COD followed by anaerobic digestion; the use of biofilm biomass (with or without carrier); and novel aeration strategies to convert NH_4^+ into NO_2^-.

2. High COD/N ratio management: strategies for maximizing C energy recovery

Municipal wastewater is a potential source of chemical energy in the form of organic carbon [17]. Besides, the COD/N ratio of this type of wastewater (typical ratios around 10–12 [18]) is usually significantly higher than the optimum desirable for a PN/Anammox treatment (<2–5, according to Lackner et al. [19], or even <0.5, according to Daigger [20]). Firstly, heterotrophs grow on biodegradable COD and compete with ammonium oxidizing bacteria (AOB) for dissolved oxygen (DO) and with Anammox for nitrite (heterotrophic denitrifiers) [19,21,22]. Secondly, if heterotrophs are growing, the production of sludge can increase and its physical characteristics can change, decreasing in that case the retention of biomass in the system [22]. Finally, some specific biodegradable organic compounds may be inhibitory for AOB [23] or Anammox biomass [7]. Regarding the outcompetition phenomena, Jenni et al. [22] have reported that the key factor for the successful operation of the process at moderate COD/N ratios (1.4 g COD/g N) is maintaining the appropriate Sludge Retention Time (SRT). Furthermore, the ability of the Anammox specie *Candidatus* "Brocadia fulgida" [24] to exhibit an organotrophic metabolism oxidizing some organic compounds (e.g. propionate, acetate, formate [24]) can increase their competitiveness at moderate COD/N ratios [22].

In any case, even when PN/Anammox could be carried out despite the COD presence, most of the biodegradable COD should be treated as a way to recover energy. As it was mentioned in the previous section, this is the main motivation to apply the PN/Anammox process in the mainstream of municipal WWTPs. Nowadays, the most practical and widely implemented way to transform biodegradable COD into recoverable energy is the anaerobic digestion process. Taking into account the stoichiometry of the methanogenesis, if the conversion is complete, the maximum production of CH_4 from biodegradable COD is about 0.35 Nm^3 CH_4/kg COD [17]. However, municipal wastewater is usually more diluted than the typical influent treated by anaerobic digestion and, consequently, its chemical energy is more difficult to harvest. The direct application of anaerobic digestion is not only hampered by this low strength but also by the moderate to low temperature of the water [17]. Except in tropical or subtropical regions, municipal wastewater would usually be 10–15°C [25]. Besides, at these moderate temperatures, a significant part of the produced CH_4, up to 40% [16], can be dissolved in the effluent, being useless in terms of energy recovery and also posing a risk of environmental release. Nevertheless, despite these drawbacks, Gao et al. [26] have successfully applied an Up-flow Anaerobic sludge Fixed-Bed (UAFB) reactor to directly treat a municipal influent prior to a PN/Anammox system. Anyway, as expected, the average COD removal was barely over 40% at 17°C. Still, this direct anaerobic treatment might be an alternative when the COD removal optimization techniques are not available.

There are some different strategies to maximize biodegradable COD recovery and its conversion into energy [14,17]. Probably, the easiest alternative to be implemented in WWTPs consists in the up-concentration of the organic carbon and the maximization of the anaerobic digestion [14,17]. This up-concentration can be performed by several techniques [16]: maximized/upgraded primary settling (Fig. 1(a) [14]), sieving, dynamic sand filtration (DSF), dissolved air flotation (DAF) or bio-flocculation/High Rate Aerobic System (HRAS) (including aerobic granulation). This last technique will aim to convert dissolved biodegradable carbon into biomass (e.g. activated sludge or aerobic granules) in high-rate/low-HRT reactors. The objective will be the maximum conversion of C into biomass, with relatively low C mineralization (i.e. conversion into CO_2) and N removal in this reactor [21]. Several authors use the name A-B stage process for all these treatments, the “A process” being the biodegradable COD concentration and methanization, and the “B process” the mainstream PN/Anammox (Fig. 1(b) [21]). Then the purged activated or granular sludge [27], together with the particulate COD, will be separated (usually by settling) and digested in order to produce biogas [14,21].

3. Low strength and low temperature wastewaters

The treatment of low strength and low temperature wastewaters, produced in such treatments like the ones discussed in the previous section to transform biodegradable COD into energy, adds difficulties not only for the Anammox step but also for the PN step. Actually, this section will focus mainly on the limitations of the PN step, because it is considered one of the main bottlenecks of this process [28].

The effective selection and growth of the AOB, outcompeting the Nitrite Oxidizing Bacteria (NOB), in order to obtain the oxidation to NO_2^- of about 50% of the NH_4^+, can be much more difficult when treating these types of wastewaters [21]. Two of the selection driving forces commonly used are based on high ammonium concentration (i.e. NOB selective inhibition by free ammonia [29,30]) and on the wash-out of NOB due to the faster growth kinetics of AOB at the mesophile range of temperature (e.g. SHARON process [31]). In this case, however, the wastewaters to be treated will be at ambient temperature, which, unless in hot/tropical climates, will be significantly lower than the mesophilic temperature range. In addition, the low ammonium concentration, usually around or under 50 mg/L [26,32], will make the inhibition by free ammonia virtually negligible [21]. In fact, Al-Omari et al. [28] reported that the out-selection of NOB can be the most challenging issue to be addressed for the effective worldwide implementation of mainstream shortcut nitrogen removal processes.

In the absence of inhibition factors to select AOB and wash-out NOB, the population selection in the PN step will have to rely on fine-tuning the concentrations of the involved species, i.e. oxygen and nitrogen species [33] and, eventually, on the use of biofilms [34]. The use of limiting DO concentrations to maintain stable conversion of ammonium to nitrite, based on oxygen affinity differences between AOB and NOB, is still a controversial matter. The main reason is that there is a wide range of oxygen affinity constants reported in the literature [35] due to the diversity of populations of AOB and NOB and also due to the different conditions of the experiments. Therefore, while some authors recommend the operation at limiting DO concentrations to supress NOB [36], others on the contrary propose the operation at non-limiting conditions [33,34]

Another alternative is described in a previous work by Bartroli et al. [37] who have focused on the measurement and control of residual ammonium concentration in the reactor and the ratio between that ammonium and the DO concentrations in order to obtain nitrification only to nitrite. They worked with high-strength wastewaters, but more recently Isanta et al. [38] have demonstrated that the same strategy can be used for municipal-like effluents (more details in one-step vs. two-step systems section and Table 1). Besides, Wett et al. [39] reported that transient anoxia (i.e. intermittent aeration) is an efficient way to control PN of municipal wastewater and repress NOB activity. They also reported that AOB bioaugmentation was beneficial for the process.

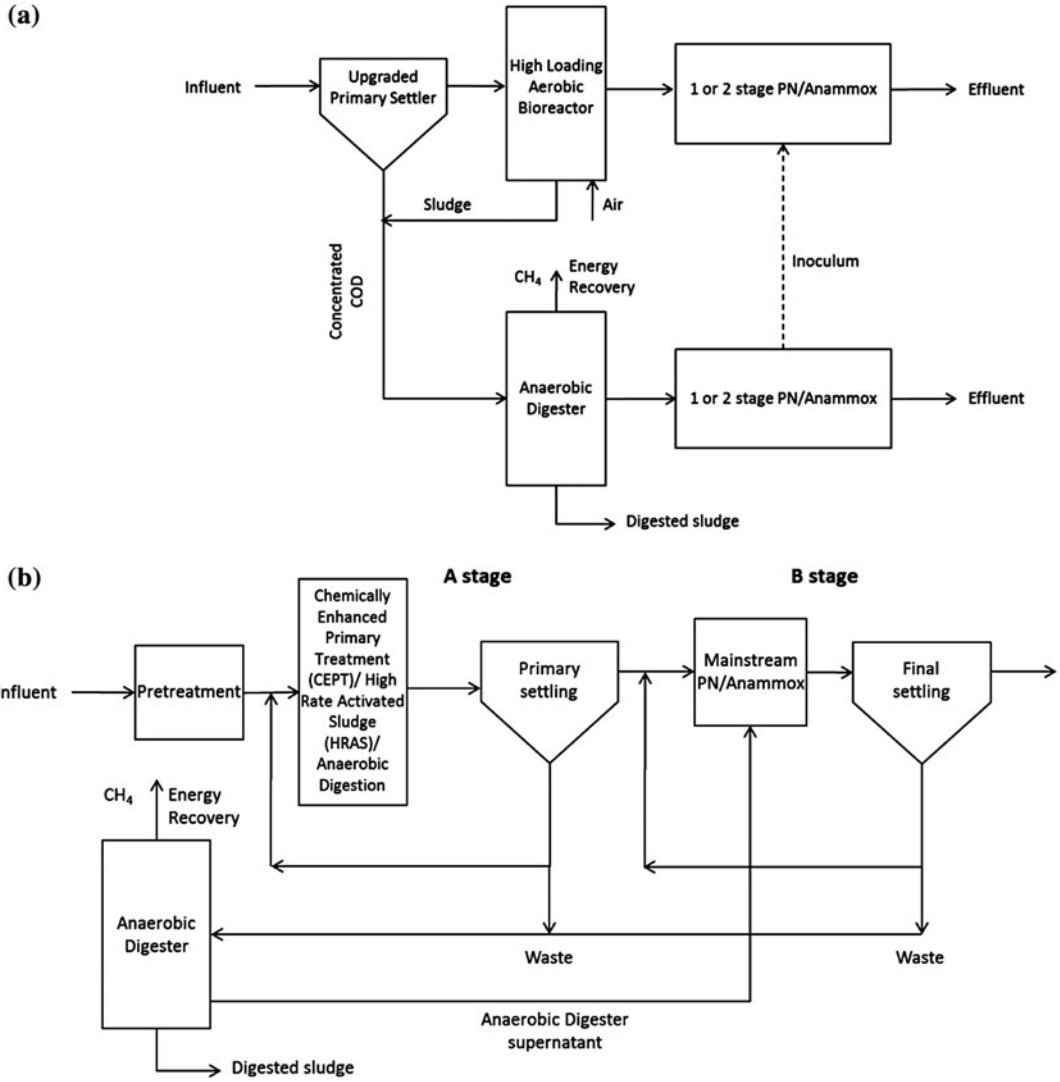

Fig. 1. (a) Adapted flow scheme of WWTP proposed by Méndez et al. [14]. Most of the biodegradable organic matter is removed in the primary settler (sedimentable COD) or converted into granular biomass in the aerobic bioreactor. Then, the primary and granular sludges are converted into biogas. A separate PN/Anammox unit in the reject water line is proposed in order to use it as an inoculum source for the PN/Anammox unit in the main line. An alternative configuration could be the direct feeding of the supernatant from the anaerobic digester to the PN/Anammox unit in the main line. (b) Flow scheme of the "A-B stage" WWTP adapted from Xu et al. [21]. Most of the biodegradable COD is concentrated in the A stage and sent to the anaerobic digester to be converted into biogas.

de Clippeleir et al. [34], however, achieved the stable operation of a one-step PN/Anammox with rotating biodisc technology treating a municipal-like synthetic influent (55–60 mg NH_4^+-N/L) at 15˚C. They needed relatively high DO concentrations (3–4 mg/L) and nitrite accumulation in the reactor (up to an average of 31% of the NH_4^+ consumed) to outcompete NOB vs. AOB and Anammox. In these conditions, the reactor produced significant amounts of undesirable nitrogen oxides. Regmi et al. [33], based on some previous works on affinity for nitrogen substrates and DO [40–43] agreed on the strategy followed by de Clippeleir et al. [34] and postulated that operating at non-limiting concentrations of NH_4^+, NO_2^- and DO, together with transient anoxia and short (or even "aggressive") SRT operation, was the best strategy to effectively control a PN system to treat municipal wastewater. Regmi et al. [33] proved their strategy by operating a pilot scale (340 L) plant under the mentioned conditions, obtaining relatively high nitrogen removal (57% on average) without carbon and alkalinity addition and at a low HRT. In order to implement

Table 1
Summary of significant experimental works on PN/Anammox treating municipal mainstream effluents

Processes	Configuration	Aeration type	COD/N ratio	Temperature (℃)	NRR (kg N/m^3 d)	Refs.
PN	Two steps: PN-CSTR, MBBR Anammox (not operated)	Intermittent, controlled by NH_4^+/NO_x ratio	6.7	25	0.4*	[33]
PN	Two steps: PN Airlift granular reactor, Anammox (not operated)	Manual control DO 1–5 mg O_2/L, continuous supply	0	12.5	0.3–0.4*	[38]
Anammox	Two steps: PN (not operated), UFGSB Anammox	–	0.6–1	10–20	1.85 (20℃) 0.34 (10℃)	[53]
Anammox	Two steps: PN (not operated), hybrid biomass Anammox SBR	–	1.8	12.5	0.05	[54]
PN/Anammox	Three steps for AD and PN/Anammox: UAFB, PN-SBR, UFBR Anammox	Aeration time controlled PN	5 (before AD)	12–27	0.83	[26]
PN/Anammox	One step: RBC	Not controlled, intermittent in space	2	14–15	0.53	[34]
PN/Anammox	One step: SBR	Not controlled, continuous supply	0	12	0.02	[25]
PN/Anammox	One step: Pilot scale plug flow granular reactor	Intermittent	0.67 (BOD/N)	19	0.16–0.19	[61]
PN/Anammox	One step: MBBR	Manual control DO < 0.3 mg O_2/L, continuous supply	0	10	0.5 g N/(kg VSS d) measured ex situ	[47]
PN/Anammox	One step: bubble column SBR	Constant DO concentration, 1.5 mg O_2/L	0.5	18	0.9	[62]
PN/Anammox	One step: SBR	Constant aeration flow rate, intermittent supply	1.4	30	0.27	[22]

Notes: AD: anaerobic digestion; PN: partial nitritation; UAFB: up-flow anaerobic sludge fixed bed; UFBR: up-flow fixed-bed biofilm reactor; SBR: sequencing batch reactor; RBC: rotating biological contactor; CSTR: continuously stirred tank reactor; MBBR: moving bed bioreactor; UFGSB: upflow fluidized granular sludge reactor; BOD: biological oxygen demand; NRR: nitrogen removal rate.
*Ammonium oxidation rate (kg N/m^3 d) is shown for PN-only systems.

the control system, the use of online sensors for nitrogen species was one of the process keys [33] and they are expected to help further to obtain even a more efficient and stable operation in future pilot or full-scale applications [28]. Actually, Al-Omari et al. [28] conducted simulation studies and concluded that the use of "ammonia vs. NOx" control was the best for mainstream nitrification. Despite all these successful works on advanced control systems, this is expected to continue being a research hotspot and the first full-scale implementations are expected in the near future.

Other aspect to be considered is that the application of PN to side-streams coming from anaerobic digestion of sludge is favoured by the fact that these types of wastewaters very often have alkalinity/ammonium molar ratios about 1–1.2 [44]. Taking into account that the oxidation of ammonium to nitrite produces two equivalents of protons per equivalent of ammonium converted, about 50–60% of the ammonium will be oxidized to nitrite when the alkalinity is totally consumed [44]. At that point, the value of pH will decrease enough to stop the PN, thus the alkalinity content of digested side-streams is helping to control the PN. The alkalinity of municipal wastewater is relatively low and alkalinity/ammonium molar ratios can easily be lower than 0.5 and, even for municipal wastewater with high alkalinity, that ratio can barely reach 0.8–0.9 [45]. This fact implies that careful control of pH is essential to perform PN process and some consumption of reagents may be necessary. However, it is also important to remark that, since the Anammox reaction consumes protons [3], when PN and Anammox are carried out in the same single-stage reactor, the alkalinity consumption caused by PN will be partially compensated by the Anammox process.

Regarding the application of the process at relatively low temperature and apart from maintaining the control/stability of the PN process, the biological activity of the Anammox population will be much lower than when relatively hot digested side-streams are treated. Despite Anammox has been proved feasible at temperatures below 20°C [8,25,45], its activity will be significantly lower (roughly 2–4 times [46]) than that observed at its optimum temperature. Even when some degree of acclimation to low temperatures is possible [8,46], the lower specific activity implies the need for high retention of Anammox biomass, topic that is discussed in the next section. Furthermore, Gilbert et al. [47] studied the influence of the type of biomass when operating at low temperatures treating municipal-like influents (50 mg NH_4^+-N/L). They observed an important impairment of Anammox activity in suspended biomass at 15°C, while it was persevered in granular biomass and in biofilms on carriers for temperatures up to <13°C. Furthermore, Anammox activity in thicker biofilms was less affected than in thinner biofilms.

4. Effective retention of Anammox biomass

Anammox bacteria grow very slowly [48,49]. There is still not a big consensus about their exact doubling time, probably in part due to the lack of a pure Anammox culture to date. Anyway, in lab-scale reactors and in conditions close to the optimum, it ranges from minimum values of 3 d [50] to 6 d [51] to a more typically reported value of about 10–12 d [9,49]. At pilot/full scale and, even more, at low temperatures the observed doubling times can go up to 25–30 d [44,52]. Furthermore, recent works on mainstream Anammox application are giving much higher values (35–123 d [53]; 18–79 d [54]). Therefore, it is clear that optimum Anammox biomass retention is essential in its application to municipal wastewater, especially at temperatures below 20–25°C.

The most widely studied and implemented mechanisms to obtain high SRT Anammox reactors rely on the formation of biofilms (either autoaggregation in form of granules or growing on support materials) [55], whose high density allows very high retention of biomass in the reaction systems. Besides, biofilms are usually more stable and less prone to suffer inhibition than suspended biomass and they allow the coexistence of several different microbial populations (e.g. Anammox and AOB). Membrane bioreactors (MBR) have also been used at lab-scale [51] and as a research tool in order to obtain total retention of biomass, but they have been barely applied at full scale. Actually, some of the recent works towards municipal wastewater treatment by PN/Anammox rely on biofilm technology [26,34,53]. Another technology, which has actually been used for Anammox reactors since its discovery [49], is the Sequencing Batch Reactor (SBR). It can be used or not in combination with biofilm systems and allows very high SRTs, close to total retention of biomass [55]. Some of last years' efforts on municipal water treatment were also successfully employing SBRs [25,33] and they are expected to continue being a popular Anammox technology in the future, not only because of good biomass retention, but also due to their versatility and operational flexibility.

5. One-step vs. two-step systems

In the case where the organic C recovery pretreatment is able to remove most of the biodegradable organic matter in wastewater, the one-step PN/Anammox process would be appropriate to carry out the subsequent ammonium removal. In fact, such a system has been proved at temperatures around 20°C removing loading rates up to 0.45 kg NH_4^+-N/(m^3 d) [56]. One of the main advantages of using one-step systems would be significantly lowering the investment costs [57], because of the savings on area occupation (and eventually purchase), civil work and equipment. Besides, as it was commented before, the consumption and production of protons of PN and Anammox can be (partially) compensated. Furthermore, the N_2O emissions associated to one-step PN/Anammox systems are generally lower (about 1% for a full-scale system operated at high ammonium concentration [58]). Most of the N_2O production is generally attributed to nitrifiers [59], so it occurs on the PN unit in the case of a system with two steps. Usually, the N_2O production by the Anammox organisms is considered as low [58,60]. Taking into account that the production by nitrifiers is significantly affected by the concentrations of NO_2^- and NH_4^+ in the reactor [59], which will be much higher in a separate PN reactor, and is also enhanced by high DO concentrations [59], the N_2O emissions will be higher in a two-step system.

If the soluble COD is not completely removed or if the variability in the influent is expected to be high, a two-step system might be more interesting, because the PN reactor can act "protecting" the Anammox unit from the COD and the changes in the influent.

For example, Gao et al. [26] chose that type of system and it was able to cope with the relatively low COD removal of the previous treatment step. Anyway, Lotti et al. [53] reported the efficient operation of a one-step Anammox reactor treating a municipal-like effluent (NLR 1.25 g N/(L d) at 15˚C/0.60 g N/(L d) at 10˚C) and receiving about 60 mg COD/L. Nevertheless, the use of two steps allows the optimization of each process, taking into account the relatively difficult control of the PN when treating this type of wastewaters. Some of the constraints and optimum values for PN and Anammox would be difficult to meet simultaneously, so with this strategy each process can be optimized independently. This is the case of the advanced PN control system proposed by Regmi et al. [33], which had to be implemented in a two-step system in order to achieve a highly efficient and stable conversion of about half the ammonium to nitrite, despite it was not specifically designed for mainstream PN/Anammox, but towards nitritation/denitritation. A later work of Isanta et al. [38] was indeed focused on mainstream PN to obtain an effluent for a subsequent Anammox treatment. They reported a stable PN at long term (300 d), operating a granular lab-scale reactor at 12.5˚C and treating an average loading rate of 0.7 g N/L d. Their control strategy was focused on assuring an adequate (i.e. very low) ratio between oxygen and ammonium concentrations in the reactor bulk liquid.

Table 1 summarizes some of the most significant examples of application of PN/Anammox to municipal wastewaters, both employing one and two-step technologies.

6. Conclusions

The application of the PN/Anammox process to the main stream of municipal WWTPs opens the possibility for the self-sufficient or energy-generating treatment plant. This highly desirable objective, both in terms of environmental sustainability and cost savings, has driven the research towards the main line implementation of the PN/Anammox. Significant advances have been obtained to overcome some of the limitations of the process: effective PN at mainstream conditions (NRR about 0.7–0.8 g N/L d); good retention of the biomass (mostly relying on granular/biofilm technologies); good performance in the presence of moderate biodegradable COD concentrations (by maintaining enough SRT and/or relying on Candidatus "Brocadia fulgida" populations) and operation at moderate to low temperatures (NRR 0.3–0.5 g N/L d at 10–15˚C). However, more work could be expected in the future, specially focused on COD concentration and C energy recovery and integration of the whole process (C management, PN and Anammox) at pilot and full scale treatment of real municipal wastewater. According to the reviewed literature and the experience of the authors, these may be the next research needs and foci. Therefore, the application of PN/Anammox to municipal wastewater is not yet a mature technology and it will continue being a hot research topic in the future.

Acknowledgements

The authors would like to acknowledge the financial support from the Spanish Ministry of Economy and Competitiveness through the project CTM2011-24897. This work was carried out within the framework of the RedNovedar (CTQ2014-51693-REDC) project. The authors also wish to acknowledge Noelia Vijande for the English proof reading.

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