**Autotrophic nitrogen removal after ureolytic phosphate precipitation to remove both endogenous and exogenous nitrogen**

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**ABSTRACT**

Anaerobic digestion yields effluents rich in ammonium and phosphate and poor in biodegradable organic carbon, thereby making them less suitable for conventional biological nitrogen and phosphorus removal. In addition, the demand for fertilizers is increasing, energy prices are rising and global phosphate reserves are declining. This requires both changes in wastewater treatment technologies and implementation of new processes. In this contribution a description is given of the combination of a ureolytic phosphate precipitation (UPP) and an autotrophic nitrogen removal (ANR) process on the anaerobic effluent of a potato processing company. The results obtained show that it is possible to recover phosphate as struvite and to remove the nitrogen with the ANR process. The ANR process was performed in either one or two reactors (partial nitritation + Anammox). The one-reactor configuration operated stably when the dissolved oxygen was kept between 0.1 and 0.35 mg L\(^{-1}\). The best results for the two-reactor system were obtained when part of the effluent of the UPP was fully nitrified in a nitritation reactor and mixed in a 3:5 volumetric ratio with untreated ammonium-containing effluent. A phosphate and nitrogen removal efficiency of respectively 83 ± 1% and of 86 ± 7% was observed during this experiment.

**Key words** | autotrophic nitrogen removal, nutrient recovery, struvite

**INTRODUCTION**

Phosphorus and nitrogen are important elements, making a major contribution to agricultural and industrial development, but their release to natural water bodies is also the main cause of eutrophication. Therefore, in Europe over the last decades wastewater treatment has been focusing on the removal of those nutrients to meet the nutrient standards according to the EU Urban Waste Water Treatment Directive 91/271/EEC (EC 1991).

Due to the population growth the demand for fertilizers is increasing. However, phosphate rock, from which phosphorus is gained, is a limited resource concerning quantity and quality (Steen 1998). On the other hand both the synthesis of ammonium from atmospheric nitrogen and the nitrification–denitrification process, as used during wastewater treatment, are energy-consuming processes with a high carbon dioxide footprint. Also fossil fuels are running out and will become more expensive in the future. Twenty years ago, scientists had already found that anaerobic digestion was the preferred technique to remove COD (chemical oxygen demand) from high-strength wastewater since this minimizes energy consumption, CO\(_2\) emission and sludge production (Six & De Baere 1992; Wilkie 2005). As a result, anaerobic pretreatment with aerobic post-treatment has already been applied in many industries instead of a direct conventional activated sludge treatment.

The global issues of nutrients (both P and N) and of food security in the future can thus only be addressed by managing nutrients in a closed cycle, in particular developing recycling and reuse of nutrients from wastewater streams. This means that besides meeting the nutrient standards, nutrient removal should be replaced by nutrient recovery or should at least be performed with a minimum
of energy consumption. This requires both changes in wastewater treatment technologies and implementation of new processes. Also the commercializing of and potential users for the recuperated nutrients are making part of these changes.

The effluent of anaerobic pretreatment systems is characterized by both the presence of varying remaining levels of COD and high nitrogen and phosphorus concentrations. Nowadays, the recovery of phosphorus is getting slowly introduced as it is a finite resource. The application of a struvite (MgNH₄PO₄·6H₂O) precipitation process in a wastewater treatment plant is beneficial to reduce the consumption of phosphate rock and to obtain a recovered product that can be used as a fertilizer or as a raw material in the phosphorus industry (de-Bashan & Bashan 2007). Ureolytic phosphate precipitation (UPP) is an example of such a struvite precipitation process (Meesschaert et al. 2007; Carballa et al. 2008; Desmidt et al. 2009). During this process an alternative way for increasing the pH is used. The pH is increased by means of bacterial urease activity, rather than by NaOH, as it occurs in the chemical process. The enzyme urease hydrolyses urea, which is added to sludge, according to the following overall reaction:

\[
\text{CO(NH}_2\text{)}_2+2\text{H}_2\text{O}\rightarrow 2\text{NH}_4^+ + \text{HCO}_3^- + \text{OH}^-
\]  (1)

Due to the release of hydroxyl ions, the pH increases during ureolysis. The process also introduces extra ammonium into the system. The addition of MgCl₂·6H₂O (2%) to the reactor, in which ammonium and hydroxyl ions are formed by ureolysis and to which both ammonium and phosphate are added with the wastewater, results in the precipitation of struvite according to the following reaction:

\[
\text{Mg}^{2+} + \text{NH}_4^+ + \text{OH}^- + \text{HPO}_4^{2-} + 5\text{H}_2\text{O} \rightarrow \text{MgNH}_4\text{PO}_4\cdot6\text{H}_2\text{O}
\]  (2)

To remove the endogenous nitrogen from the wastewater and exogenous nitrogen, introduced by the urea, an additional treatment step is needed. A recently developed process to treat nitrogen-loaded wastewaters is the autotrophic nitrogen removal process (ANR). The ANR process is a combination of a partial nitritation and an Anammox (anaerobic ammonium oxidation) reaction.

In view of the coupling of partial nitritation with Anammox, nitrite-oxidizing activity should be suppressed and ammonium should only be oxidized by about 50% to nitrite (van Dongen et al. 2001):

\[
\text{NH}_4^+ + 0.75\text{O}_2^- \rightarrow 0.5\text{NO}_2^- + 0.5\text{NH}_4^+ + \text{H}^+ + 0.5\text{H}_2\text{O}
\]  (3)

A number of strategies have been suggested for the wash-out of nitrite-oxidizing bacteria (NOB) to achieve partial nitritation such as elevated temperature (30–40 °C), a dilution rate of 1–2 days and low DO (dissolved oxygen) (Blackburne et al. 2008). A 50% conversion of NH₄⁺ to NO₂⁻ can be obtained when the wastewater has a neutral pH and a molar TIC:TAN (total inorganic carbon:total ammonium nitrogen) ratio of 1:1. In that case, the amount of protons produced during conversion of the ammonium to nitrite is equal to the amount of protons taken up via carbon dioxide stripping. Hence, ammonium oxidation will stop at 50% conversion due to acidification, and an Anammox-suited influent is produced (van Dongen et al. 2001). Anammox bacteria are able to consume the remaining ammonium and previously formed nitrite (from the partial nitritation) under anaerobic conditions according to the following reaction (Strous 1998; Pynaert et al. 2004):

\[
\text{NH}_4^+ + 1.32\text{NO}_2^- \rightarrow 1.02\text{N}_2 + 0.26\text{NO}_3^- + 2.03\text{H}_2\text{O}
\]  (4)

A small fraction of nitrite is thus anaerobically oxidized to nitrate and yields electrons for the reduction of carbon dioxide to cell material for cell growth.

Compared with the conventional nitrification–denitrification process, the ANR process consumes 65% less oxygen and 100% less biodegradable organic carbon, resulting in a lower operating cost (Verstraete & Philips 1998). In order to implement the ANR process it is important to take into account that the process can be performed in two different units or in a single one. This latter process is designated as CANON (completely autotrophic nitrogen removal over nitrite, Sliekers et al. 2002), OLAND (oxygen-limited autotrophic nitrification–denitrification, Kui & Verstraete 1998) or DEMON (DEamMONiFication process, Wett et al. 2007). Under oxygen-limited conditions a co-culture of aerobic and anaerobic ammonium oxidizing bacteria (AnAOB) can be established in a single unit. The overall N removal stoichiometry (Pynaert et al. 2004) for those processes can be given as:

\[
\text{NH}_4^+ + 0.85\text{O}_2 \rightarrow 0.11\text{NO}_3^- + 0.44\text{N}_2 + 1.44\text{H}_2\text{O} + 1.14\text{H}^+
\]  (5)
In those systems, NOB compete for oxygen with the aerobic ammonium oxidizing bacteria (AerAOB) and for the nitrite with Anammox bacteria, and thus its growth (and subsequent nitrate production) is prevented. Another reason to maintain low oxygen concentrations is that Anammox bacteria are reversibly inhibited by DO concentration higher than 0.5% of air saturation (Strous et al. 1999). The main disadvantage of these processes is their reliance on the low growth rate of Anammox bacteria. To enhance the performance of reactors involving slow-growing bacteria, high sludge retention times are mandatory, and therefore the attachment of bacteria on a carrier material to develop biofilms or the self-aggregation concept in granules are being applied.

The aim of this work was to investigate if the ANR process could remove both the endogenous ammonium from the effluent of an UASB (upflow anaerobic sludge blanket) and the ammonium derived from the ureolytic struvite precipitation. For the ANR process both the one- and the two-unit configurations were tested. In the two-unit configuration the partial nitritation process was carried out in a continuous stirred tank reactor (CSTR). Once an adequate nitrite:ammonium ratio was obtained, the effluent of the CSTR was fed to an Anammox reactor with carrier material. For the study of the single-unit configuration, the previously mentioned Anammox reactor was used, which became gradually inoculated with sludge of the partial nitritation process and as such provoked a change in the ANR from a two-reactor configuration to a one-reactor strategy. The name OLAND originated from Ghent University (Kuai & Verstraete 1998) and will be used throughout this paper for the one-reactor configuration.

**MATERIALS AND METHODS**

**Experimental set-up**

**Ureolytic phosphate precipitation**

Based on previous experiences at pilot scale, further experiments were carried out at laboratory scale on anaerobic effluent of a potato processing company (Agristo NV, Harelbeke, Belgium). The reactor at laboratory scale (built according to BNB EN ISO 11733 (ISO 2004); working volume of 4 L) is a simplified configuration without air stripper. Previous experiments at pilot scale (Desmidt et al. 2012) have shown that the average pH after air stripping was 7.8. Instead of first increasing the pH of the anaerobic effluent by air stripping, the pH of the anaerobic wastewater was increased to 7.8 with NaOH before it was fed to the crystallization reactor, where struvite precipitation occurred. Figure 1 shows a schematic overview of the configuration used at laboratory scale.

During operation, urea (1.5%) was added to the crystallization reactor, in which anaerobic sludge was present. The hydraulic retention time (HRT) of the wastewater in the crystallization reactor varied between 6 and 7 hours.

The effluent from the crystallization reactor was directed to a decanter, where separation between the final effluent and sludge and struvite occurred. The struvite was sluiced from the reactor (Figure 1).

**Autotrophic nitrogen removal: two-reactor configuration (partial nitritation and Anammox)**

After decantation, the effluent of the UPP was fed to a continuous reactor of 20 L, inoculated with nitrifying sludge of a potato processing company (Agristo NV, Harelbeke, Belgium).

The operational conditions during the partial nitritation process for washing out the NOBs were: no sludge retention and a temperature of 35 °C. Since the dilution rate depends on the influent ammonium concentration and the pH, the HRT was estimated according to Volcke et al. (2007). No pH adjustment occurred in the reactor. At the start of the experiment, the DO was controlled between 3 and 4 mg L⁻¹. At stable operation, the DO was gradually lowered to 0.5 mg L⁻¹.

The Anammox bacteria in the second reactor were enriched from conventional sludge of a vegetable processing company (Unifrost NV, Koolskamp, Belgium) in a
A laboratory-scale reactor with a volume of 3.7 L and filled with carrier material which consisted of polyurethane. The reactor operated as an upflow fixed bed reactor (UFBR). In the middle of the carrier material a glass tube was provided, making mixing and feeding at the bottom of the reactor possible. The effluent was drained off at the upper side of the reactor. The underpart of the reactor was continuously mixed with a mechanical stirrer and the reactor was kept at a temperature of 35°C. To enrich the Anammox bacteria the reactor was first fed with synthetic medium. The composition of the medium is described in Appendix 1 (available online at http://www.iwaponline.com/wst/067/666.pdf).

The synthetic medium was fed to the reactor with a HRT of 1.6 days. After 3 months of operation, the Anammox reactor was fed with part of the effluent of the partial nitritation reactor. Due to the slow growth rate of the Anammox bacteria and thus the low removal capacity of the Anammox reactor, the effluent of the partial nitritation was fed to the Anammox reactor at a volume ratio of 1:6. Figure 2 shows a simplified scheme of the ANR in two reactors.

**Autotrophic nitrogen removal: OLAND configuration**

By feeding the Anammox reactor with the effluent of the partial nitritation (without sludge retention), some sludge of the partial nitritation got caught in the carrier material of the Anammox reactor leading to a co-culture of microorganisms. After 2 months of operation in the two-reactor configuration, the original Anammox reactor was used as an OLAND reactor.

**Autotrophic nitrogen removal: up-scaling**

The configuration which gave the best nitrogen removal efficiency was tested on a larger scale in order to have a continuous process. Therefore an Anammox reactor with a working volume of 20 L was used. This Anammox reactor was started up with sludge of a manure processing company (DANIS NV, Izegem, Belgium) and was also inoculated with sludge obtained from a laboratory-scale rotating biological contactor with OLAND activity (LabMET, University of Ghent, Belgium, Kuai & Verstraete 1998). Before the test the 20 L Anammox reactor was also fed with synthetic wastewater according to Appendix 1.

**Calculations**

The AnAOB activity, NOB activity and AerAOB activity were estimated based on nitrogen balances and stoichiometry according to Equations (6)-(9) (Vazquez et al. 2009). The AnAOB activity includes the total nitrogen that is removed and also the 26% (Equation (3)) or 11% (Equation (4)) of NH4+-N that is consumed for autotrophic NO3-N formation in the two-reactor configuration and OLAND configuration respectively. The NOB activity includes the remainder of the NO3-N production, thus the real nitrification. The AerAOB activity is the sum of the activity that leads to the nitrite that is used by the NOBs, to the nitrite that is used by the AnAOB and to the excessive nitrite production.

\[ \Delta N = (NH_4^+ - N_{influent} - NH_4^- - N_{effluent}) + (NO_2^- - N_{effluent} - NO_2^- - N_{influent}) + (NO_3^- - N_{effluent} - NO_3^- - N_{influent}) \]  
\[ \text{AnAOB activity (mg N.L}^{-1}.d}^{-1} = \frac{\Delta N}{\text{HRT}} + \frac{x}{y} \frac{\Delta N}{\text{HRT}} \]  
\[ \text{NOB activity (mg N.L}^{-1}.d}^{-1} = \frac{(NO_3^- - N_{effluent} - NO_3^- - N_{influent})}{\text{HRT}} \]  

Figure 2 | Simplified scheme of the ANR in two reactors (adapted and modified from van Dongen et al. (2001)) given with relative amounts of ammonium.
AerAOB – activity (mg N L\(^{-1}\) d\(^{-1}\))

\[
\begin{align*}
\text{AerAOB} & = \frac{(\text{NO}_3^- - \text{N}_{\text{effluent}} - \text{NO}_3^- - \text{N}_{\text{influent}}) \cdot \frac{x \cdot \Delta N}{\text{HRT}} + z \left( \frac{\Delta N}{\text{HRT}} + \frac{x \cdot \Delta N}{y \cdot \text{HRT}} \right) + (\text{NO}_2^- - \text{N}_{\text{effluent}} - \text{NO}_2^- - \text{N}_{\text{influent}})}{\text{HRT}}
\end{align*}
\]

with \(x\), \(y\) and \(z\) equal to 0.11, 0.88 and 1.32/2.32 for an OLAND configuration and 0.26, 2.04 and 0 for a two-reactor configuration.

Analytical methods

The pH was measured with a pH meter (Mettler Toledo seven multi). DO was measured with an oxygen meter Oxi 315 (WTW). Ammonium and magnesium were determined with a Dionex DX-100 chromatograph equipped with a conductivity detector. Phosphate, nitrite and nitrate were measured with a Shimadzu total carbon analyzer TOC-ductivity detector. Inorganic carbon (IC) was analyzed by a detector. Phosphate, nitrite and nitrate were measured with a Dionex series 4500i ion chromatograph equipped with a conductivity detector. Inorganic carbon (IC) was analyzed by a Shimadzu total carbon analyzer TOC-VC. The crystals formed in the ureolytic phosphate reactor were examined and identified by X-ray diffraction (XRD), using a Stoe Stadi P unit with Cu \(K\alpha\) radiation at 0.1541 nm with an image plate as detector. The DO in the partial nitritation reactor was measured by means of an LDO (luminescent dissolved oxygen) electrode coupled with an SC-100 controller of Hach-Lange.

Fluorescent in situ hybridization (FISH)

The biomass was fixed in a 4% paraformaldehyde solution and FISH was performed according to Amann et al. (1990). The probes used in this study were Nso1225 labeled with fluorescein for b-proteobacterial AOB (Mobarry et al. 1996) combined with Amx820 labeled with Cy3 for the Anammox bacteria ‘Candidatus Brocadia’ and ‘Candidatus Kuenenia’ (Schmid et al. 2000). Image acquisition was done on a Zeiss Axioskop 2 Plus epifluorescence microscope.

RESULTS AND DISCUSSION

Ureolytic phosphate precipitation

For practical reasons and to investigate if the process still obtained a good phosphate removal efficiency at lower phosphate concentrations, the anaerobic effluent of the company was diluted two times with demineralized water. Table 1 shows the characteristics of the influent and effluent of the UPP. The hydrolysis of urea resulted in a pH increase and an increase in ammonium and IC concentration.

The TIC:TAN ratio decreased from 2.30 ± 0.08 to 1.88 ± 0.02. The TIC:TAN ratio thus decreased after UPP. The TIC:TAN ratio is, however, still higher than 1, which is higher than the generally tested ratio to obtain a 50% conversion of \(\text{NH}_4^+\) to \(\text{NO}_2^-\).

The addition of magnesium chloride to the reactor resulted in a higher magnesium concentration in the effluent. During the experiments a high phosphate removal efficiency of 85 ± 1% was observed, resulting in a final effluent concentration of \(8 \pm 3 \text{ mg L}^{-1}\) PO\(_4\)-P. The crystals in the reactor were identified as struvite by XRD (Appendix 2, available online at http://www.iwaponline.com/wst/067/666.pdf).

Autotrophic nitrogen removal: two-reactor configuration

After the phosphate precipitation the ammonium was removed by a two-reactor configuration, existing of a combination of a partial nitritation and Anammox process. Good operation of a partial nitritation reactor is achieved by applying a dilution rate that is high enough to wash out nitrite oxidizers, but low enough to ensure growth of aerobic ammonium oxidizers. According to Volcke et al. (2007) the dilution rate depends on the influent ammonium concentration and the pH. Based on the average effluent ammonium concentration (460 mg L\(^{-1}\) NH\(_4^+\)), the average effluent pH (pH 8.04) as observed in our previous experiments at pilot scale (Desmidt et al. 2012) and the fact that the wastewater was diluted twice with demineralized water at laboratory scale, we calculated the dilution rates that guaranteed a stable nitrite formation. These dilution rates lie between 0.5 and 1.25 d\(^{-1}\). So, a HRT of 1.4 days was a good choice to ensure robust operation. Volcke et al. (2002) also found that the TNO\(_2\):TAN (total nitrite

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Influent UPP</th>
<th>Effluent UPP</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH</td>
<td>7.9 ± 0.2</td>
<td>8.3 ± 0.1</td>
</tr>
<tr>
<td>PO(_4)-P</td>
<td>57 ± 10</td>
<td>8 ± 3</td>
</tr>
<tr>
<td>NH(_4)-N</td>
<td>203 ± 56</td>
<td>261 ± 54</td>
</tr>
<tr>
<td>Mg(^{2+})</td>
<td>39 ± 14</td>
<td>155 ± 35</td>
</tr>
<tr>
<td>IC</td>
<td>396 ± 96</td>
<td>421 ± 91</td>
</tr>
</tbody>
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nitrification ratio in the effluent of the partial nitritation process is highly influenced by the buffer capacity of the influent, which varies with influent pH and TIC concentration. The influent pH and the average TIC:TAN ratio in the influent of our experiments were 8.3 and 1.88 respectively. As a consequence the partial nitritation of this type of wastewater can go further than the ideal TNO2:TAN ratio of 1:1

Figure 3 shows that a continuous aeration of the reactor resulted in almost a complete conversion of ammonium to nitrite, even at low DO concentrations (0.5–1 mg L⁻¹). Ruiz et al. (2003) studied the behavior of the nitrification system during consecutive changes in DO values. The results showed that no nitrite accumulated at DO values of 5.7–2.7 mg L⁻¹. At a DO of 1.4 and 0.7 mg L⁻¹ nitrite accumulation increased with the same ammonia consumption, and at a DO of 0.5 mg L⁻¹ both nitrite accumulation and ammonia consumption decreased. Therefore, the DO was kept at 0.5 mg L⁻¹ in the last period of our experiment. During this period ammonium and nitrite were simultaneously present (Figure 3, day 100–140): a more or less stable conversion of 50% NH₄-N to NO₂-N was observed. During this period the effluent of the partial nitritation reaction between days 100 and 140 was thus the influent of the Anammox reactor between days 1 and 40.

The results of the Anammox reactor are given in Figure 4. Figure 4(a) shows the nitrogen loading rate and the nitrogen removal rate. The percentage of NO₃-N produced per NH₄-N consumed and the N removal efficiency are shown in Figure 4(b). The AnAOB activity, NOB activity and AerAOB activity were calculated according to Equations (6)–(9) and are shown in Figure 4(c). The presence of Anammox bacteria in the Anammox reactor was confirmed by FISH analysis (Appendix 3, available online at http://www.iwaponline.com/wst/067/666.pdf).

During the first 20 days of operation, the nitrogen removal efficiency was 79 ± 7% and resulted in a final effluent concentration of 31 ± 10 mg L⁻¹ NH₄-N. During the following weeks the removal efficiency decreased. The decrease in removal efficiency was probably due to the NO₂-N:NH₄-N molar ratio in the effluent of the partial nitritation which varied along the period. From Figure 3 it can be calculated that the NO₂-N:NH₄-N ratio in the effluent of the partial nitritation process between day 120 and 130 was sometimes higher than the theoretical molar ratio of 1.32 (Equation (3)). This variation resulted in a build-up of the nitrite concentration in the effluent of the Anammox reactor and subsequently in a decrease of the Anammox activity (AnAOB activity), as can be seen in Figure 4(c) from day 20 to 30. When the NO₂-N:NH₄-N ratio was again around 1.32 or lower (Figure 3, day 130–140), the Anammox activity increased again (day 30–40) and subsequently so did the nitrogen removal efficiency (Figure 4(b)). From these results it can be concluded that controlling the DO at 0.5 mg L⁻¹ in the reactor was not sufficient to obtain a continuous stable nitrite:ammonium ratio in the effluent of the partial nitritation reactor. This was related to the TIC:TAN ratio (1.88 ± 0.02) of the wastewater, which was higher than 1, and consequently resulted in an unstable TNO₂:TAN ratio in the effluent of the partial nitritation reactor.

Figure 3 | Percentage of NO₂⁻N (●), NO₃⁻N (▲) and NH₄-N (△) during partial nitritation of ammonium at different DO concentrations.
Autotrophic nitrogen removal: OLAND configuration

From day 30 to day 40 (Figure 4(c), period 1) an increase in AerAOB activity and a decrease in nitrate production (Figure 4(b), period 1) were noticed. This was indicative for a partial nitritation activity in the Anammox reactor. By feeding the Anammox reactor with the effluent of the partial nitritation reactor (without sludge retention), some
sludge of the partial nitration apparently was caught in the carrier material of the Anammox reactor leading to a co-culture of micro-organisms. To test this hypothesis, the Anammox reactor was fed directly with the effluent of the UPP instead of with the effluent of the partial nitration reactor. We thus tested if it was possible to operate the ANR process as OLAND instead of with the two-reactor configuration. From day 45 to day 135 (Figure 4, period 2) the DO was kept between 0.1 and 0.35 mg L\(^{-1}\). On day 138 the DO was increased and kept between 0.1 and 0.5 mg L\(^{-1}\). From that moment on the NOB activity increased (Figure 4(c)). As a consequence the NO\(_3\)-N concentration increased in the effluent (Figure 4(b)). The best operational DO for the OLAND configuration thus lay between 0.1 and 0.35 mg L\(^{-1}\). The AnAOB activity in period 2 of the experiment was lower than in period 1 (Figure 4(c)), leading to a lower N removal efficiency (Figure 4(b)).

**Autotrophic nitrogen removal: up-scaling**

Figure 4(b) shows that the highest nitrogen removal ratios were obtained with the two-reactor configuration in the first period (day 1–20) when the system worked under lower NO\(_2\) concentrations. Therefore this configuration was retested on an Anammox reactor of 20 L. As described earlier, the Anammox conversion efficiency decreases rapidly when the nitrite:ammonium ratio in the Anammox feed deviates from the ideal ratio. Therefore controlling the nitrite:ammonium ratio in a two-reactor configuration is essential to avoid toxic nitrite concentrations, which inhibit the Anammox conversion. Figure 3 shows that aeration between 0.5 and 1 mg L\(^{-1}\) resulted in a 90% conversion of NH\(_4\)-N to NO\(_2\)-N. This high conversion was attributed to the fact that the TIC:TAN ratio of the wastewater was 1.88, and thus the TIC concentration was sufficient to take up 90% of the protons formed during the oxidation of ammonium to nitrite (reaction 2). Instead of lowering the DO to 0.5 mg L\(^{-1}\) to obtain a 50% conversion of NH\(_4\)-N to NO\(_2\)-N, another strategy was tested. The ammonium was fully oxidized (operating DO: 0.5–1 mg L\(^{-1}\)) to nitrite in part (3:5) of the effluent of the UPP and then mixed with the remaining effluent before feeding it to the Anammox reactor. In this way the influent of the Anammox reactor had a stable 50–50% ratio of NH\(_4\)-N to NO\(_2\)-N.

The results of the 20 L Anammox reactor are also shown in Figure 4 (period 3). Compared with period 1 of the experiment, the 20 L reactor had a higher Anammox activity (Figure 4(c)) which corresponded to 172 ± 9 mg N L\(^{-1}\) d\(^{-1}\). This activity was high enough to process all the wastewater after the UPP. Also the Anammox activity was more stable during this third period than in the first period of the experiment. This was addressed to the more stable composition of the wastewater which was fed to the Anammox reactor. During this third period an average nitrogen removal efficiency of 86 ± 7% was observed. In Table 1 it is shown that, during UPP, the ammonium concentration increased from 205 ± 56 to 261 ± 54 mg L\(^{-1}\). As the system removes 86 ± 7% of the total nitrogen, it is clear that both endogenous ammonium, which was already present in the UASB effluent, and the exogenous ammonium, which is introduced during the UPP, were largely removed. The percentage of nitrate that was produced per ammonium consumed varied between 9 and 15%, which is lower than the theoretical 26% (Equation (3)). When the amount of nitrate formed is expressed as the percentage of the total nitrogen treated by the system, it is 3.9–6.5%. The reduction of COD from 260 to 100 mg L\(^{-1}\) indicated that the process went in combination with some denitrification and explained the lower nitrate concentration in the effluent.

**CONCLUSIONS**

The results obtained during this work show that it is possible to recover phosphate as struvite and to remove both the endogenous and exogenous nitrogen with the autotrophic nitrogen process from wastewater after anaerobic digestion coming from a potato processing company. For the OLAND configuration stable results were obtained when the DO was kept between 0.1 and 0.35 mg L\(^{-1}\). In the conditions tested an average N removal efficiency of 60% was obtained. Higher Anammox activity would be necessary to obtain a higher N removal efficiency.

For the implementation of a two-step ANR process on wastewater with a TIC:TAN ratio higher than 1, a configuration is proposed in which the full buffering capacity of TIC is exploited for the maximal oxidation of ammonium to nitrite in part of the wastewater to be treated. In our case (TIC:TAN = 1.88 ± 0.02) stable results were obtained when three-fifths of the effluent of the UPP was fully nitrified in the nitritation reactor (operating DO: 0.5–1 mg L\(^{-1}\)) before being mixed with the remainder of the effluent and treated in the Anammox reactor. This combination of the UPP and the ANR process in the two-reactor configuration resulted in a
phosphate and nitrogen removal efficiency of respectively 83 ± 1% and of 86 ± 7%.

REFERENCES


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