

# Evaluation of nitrous oxide gaseous emissions from a partial nitritation reactor operating under different conditions

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#### ABSTRACT

In this study, a laboratory-scale partial nitritation (PN) reactor was fed with a synthetic medium simulating the ammonium-rich wastewater produced by the anaerobic digestion of food waste. The reactor was operated at constant hydraulic retention time (1 d) and nitrogen loading rate (1.5 g N/L d), with different influent alkalinity to ammonium-nitrogen molar ratios (Alk/N, 1 and 1.3) and dissolved oxygen (DO) concentrations (5.0, 3.0, 2.0, 1.5, and  $1.0 \text{ mg O}_2/L$ ). For each operating condition tested, nitrous oxide (N,O) gaseous emissions from the PN reactor were measured via infrared gas-filter correlation, in order to achieve a deeper understanding of process potential environmental impact. As DO concentration ranged between 5.0 and 1.5 mg O<sub>2</sub>/L, the partial conversion of ammonium to nitrite was successfully achieved, with negligible nitrate production and nitrous oxide emission. When Alk/N was increased from 1 to 1.3 (DO was not limiting), the increase in ammonium-nitrogen oxidation rate (AOR, from  $717 \pm 17$  to  $945 \pm 21$  mg NH<sub>4</sub>-N/L d) and the simultaneous decrease in N<sub>2</sub>O-N emission factor (from  $0.33\% \pm 0.01\%$  to  $0.23\% \pm 0.01\%$  of AOR) were observed. When DO was set to  $1.0 \text{ mg O}_2/\text{L}$ , PN was irreversibly compromised, and a corresponding increase in N<sub>2</sub>O-N emission factor was observed (from  $0.22\% \pm 0.01\%$  to  $0.61\% \pm 0.03\%$  of AOR). Nitrifier denitrification was suggested as the main pathway contributing to N<sub>2</sub>O emission. Minimization of anoxic conditions may contribute in reducing greenhouse gas emissions even at low DO concentrations, as long as they are not process-limiting.

Keywords: Alkalinity; Ammonium; Greenhouse gas emissions; Nitrous oxide; Partial nitritation

# 1. Introduction

According to the intergovernmental panel on climate change [1], nitrous oxide ( $N_2O$ ) has a strong ozone layer depleting potential, and the third largest radiative forcing (i.e., the capacity of a gas to affect the balance between incoming solar radiation and outgoing infrared radiation, thereby contributing to climate change) among the anthropogenic gases, with an estimated lifetime of 131 years and a global warming potential up to 300-fold higher than  $CO_2$ .

Although the emissions of nitrous oxide from wastewater treatment plants (WWTPs) are relatively small (3% of the estimated total anthropogenic N<sub>2</sub>O emissions), indeed they represent a significant factor (26%) in the greenhouse gas footprint of the total water chain [2]. In recent years, the assessment of N<sub>2</sub>O emissions from WWTPs, in particular from biological nitrogen removal processes, has become of great environmental concern. Law et al. [3] reported two key metabolic pathways involved in N<sub>2</sub>O production by autotrophic ammonia oxidizing bacteria (AOB): autotrophic ammonia oxidation, where N<sub>2</sub>O can be formed as a side product during the conversion of the ammonium-oxidation intermediate hydroxylamine (NH<sub>2</sub>OH) to nitrite; and nitrifier

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denitrification (i.e., the reduction of  $NO_2$  to NO and  $N_2O$  by autotrophic AOB, under oxygen limiting conditions).

For the treatment of ammonium-rich liquid streams like, among the others, reject water, landfill leachate, livestock manure, and petrochemical wastewater [4–7], the combination of partial nitritation (PN) and anammox (ANaerobic AMMonium OXidation) has been proved to be an efficient and cost-effective solution, compared with conventional biological processes based on nitrification and denitrification. However, shortcut nitrogen removal via the nitrite pathway is likely a major contributor to overall  $N_2O$  emission [8], since nitrite is reported to trigger nitrous oxide production [2].

In this study, a laboratory-scale PN reactor was fed with an ammonium-rich synthetic medium simulating the  $NH_4$ -N content and alkalinity of the liquid effluent produced by the anaerobic digestion of food waste (AD-FW), and gaseous  $N_2O$  emissions were measured with different operating conditions, in order to strike the right balance between overall process performance and  $N_2O$  release in the atmosphere.

#### 2. Materials and methods

#### 2.1. Reactor setup and operation

The PN unit consisted of a 2 L continuous flow stirred tank reactor operated as a chemostat (without biomass recirculation). A thermostatic bath was used to control temperature at 35°C  $\pm$  0.5°C; pH was constantly monitored and kept within the range 6.0–7.5 by dosing acid (H<sub>2</sub>SO<sub>4</sub>/ 1 M) or base (NaOH, 1 M) solutions.

During the experiments, dissolved oxygen (DO) concentration was continuously monitored, and maintained at the desired level by supplying a variable mixture of air and dinitrogen gas in the bulk liquid at a constant rate (1 L/min).

A schematic representation of the experimental apparatus is reported in Fig. 1.

- 1. Sealed reactor vessel
- 2. DO, pH and temperature probes
- 3. Stirrer
- 4. Thermostatic bath
- 5. DO, pH and temperature data aquisition and control system
- 6. Solenoid valve
- 7. Mixing chamber
- 8. Gas flow meters
- 9. Influent line and pump
- 10. Effluent line and pump
- 11. Gas conditioner
- 12. In line gas analyzer and data logger

As reported in Milia et al. [9], the reactor was inoculated with activated sludge drawn from the municipal WWTP of Cagliari (Italy), fed with an ammonium-rich synthetic medium (1,500 mg  $NH_4$ -N/L), and operated for 4 months before carrying out the experiments described in this study.

Influent flow rate was kept at 1.4 mL/min, resulting in a hydraulic retention time (HRT) and a corresponding sludge retention time (SRT) of 1 d. Total nitrogen loading rate (NLR) was 1.5 g N/L d. The influent alkalinity to ammoniumnitrogen molar ratio (Alk/N) was increased from 1 to 1.3 by adding bicarbonate (as NaHCO<sub>3</sub>) to the medium. The plan of the experimental activity is summarized in Table 1.

The overall composition of the synthetic medium was:  $NH_4HCO_3 8,466 mg/L$ ,  $KH_2PO_4 1,000 mg/L$ ,  $MgSO_4 100 mg/L$ ,  $NaHCO_3 0-2,700 mg/L$ , and 10 mL/L trace elements according to Milia et al. [6]. The resulting pH was 7.8–8.0.

### 2.2. Analytical procedures

Influent and effluent NH<sub>4</sub>-N concentration was determined according to Standard Methods [10], using a Hitachi U-2000 spectrophotometer at a wavelength of 420 nm.

# Table 1Plan of the experimental activity

Phase	Duration (d)	DO (mg O <sub>2</sub> /L)	Alk/N (–)
1	34	5.0	1.0 <sup>a</sup>
2	19	3.0	1.0
3	17	2.0	1.0
4	21	2.0	1.3 <sup>b</sup>
5	21	1.5	1.3
6	6	1.0	1.3

<sup>a</sup>Typical value reported in literature for synthetic influents fed to PN reactors.

<sup>b</sup>Average value observed in real AD-FW wastewater.



Fig. 1. Schematic representation of the experimental apparatus.

The concentration of NO<sub>2</sub>-N and NO<sub>3</sub>-N was determined by ion chromatography using a DIONEX ICS-90 equipped with an AS14A Ion-PAC 5  $\mu$ m column. All samples were filtered (0.45  $\mu$ m) before analyses, which were performed in triplicate. Free ammonia (FA) and free nitrous acid (FNA) concentrations were estimated according to Anthonisen et al. [11]. Total suspended solids and volatile suspended solids (VSS) concentrations were determined according to Standard Methods [10].

Nitrous oxide measurement campaigns were carried out during each experimental phase, as steady state conditions were achieved (the only exception was Phase 6, due to the lack of process stability): headspace gas was collected from the reactor at constant flow rate (1 L/min), sent to a gas conditioning system (Bühler, mod. TGAK 3) and then to a gas analyzer (Servomex, mod. 4100), where continuous measurement of N<sub>2</sub>O concentration (as ppmv) was performed via infrared gas-filter correlation. Continuous mixing and aeration avoided N<sub>2</sub>O accumulation in the bulk liquid. Each measurement campaign lasted from 13 to 24 h, and data acquisition rate was set at 1 sample/min. The N<sub>2</sub>O-N emission rate (ER) was calculated according to Lv et al. [12], with some modifications (Eq. 1) as follows:

$$ER = \frac{c \times Q \times p \times M_N \times 2}{(1,000 \times R \times T \times V_L)}$$
(1)

where ER is the N<sub>2</sub>O-N emission rate (mg N/L d), *c* is the N<sub>2</sub>O level in the gas sample (ppmv), *Q* is the volumetric flow rate of the off-gas (L/d), *p* is the atmospheric pressure (1 atm),  $M_N$  is the molar mass of nitrogen (g/mol), *R* is the gas constant (0.082056 L atm/mol K), *T* is the temperature (K), and  $V_L$  is the working volume of the reactor (L).

The  $N_2$ O-N emission factor (EF) was then calculated by dividing ER by the corresponding average ammoniumnitrogen oxidation rate (AOR, mg N/L d). Liquid samples were also collected from the mixed liquor at the beginning/end of each  $N_2O$  measurement campaign, and analyzed in order to determine  $NH_4$ -N,  $NO_2$ -N, and  $NO_3$ -N concentrations. The dissolved  $N_2O$  concentration could not be measured in the present experiment; by the way, continuous blowing of a variable mixture of air and dinitrogen gas at a constant rate allowed continuous stripping of dissolved nitrous oxide from the liquid phase. Moreover, given the operational conditions applied, gas solubility could be considered as a constant, thus, the evaluation of nitrous oxide gaseous emissions could be assumed as representative of overall  $N_2O$  production.

# 3. Results and discussion

#### 3.1. Reactor performance

During the whole experiment, error in nitrogen balances averaged out at less than 1%. As shown in Fig. 2, the decrease in DO concentration from 5 to 2 mg O<sub>2</sub>/L (Phases 1-3) did not cause any significant change in NH<sub>4</sub>-N, NO<sub>2</sub>-N, and NO<sub>3</sub>-N effluent concentrations; coherently, the average ammonium removal efficiency and effluent NO<sub>2</sub>/NH<sub>4</sub> molar ratio measured through Phases 1-3 were 47% ± 3% and  $0.9 \pm 0.1$ , respectively, indicating very stable process performance. Despite the high process stability, such values were found to be slightly lower than expected (i.e., 50% and 1.0, respectively), considering the applied Alk/N molar ratio [13]. As suggested by Van Hulle et al. [13], the short applied HRT (1 d) may have caused a mild limiting effect on AOB cell growth, as confirmed by the relatively low biomass concentration achieved in the PN reactor (Table 2). As for other possible inhibiting factors, such as an excess of FA and FNA, or a lack of inorganic carbon, they can be reasonably excluded, since they were outside the ranges considered as inhibiting, according to Van Hulle et al. [14] and Guisasola et al. [15], respectively.



Fig. 2. Trends of  $NH_4$ -N (influent and effluent),  $NO_2$ -N (effluent), and  $NO_3$ -N (effluent) concentrations observed during the whole experimental study.

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Phase	Influent NH <sub>4</sub> -N (mg/L)	Effluent NH <sub>4</sub> -N (mg/L)	Effluent NO <sub>2</sub> -N (mg/L)	Effluent NO <sub>3</sub> -N (mg/L)	NH <sub>4</sub> removal efficiency (%)	Effluent NO <sub>2</sub> /NH <sub>4</sub> ratio (–)	Biomass concentration (mg VSS/L)	Maximum FA concentration (mg NH <sub>3</sub> /L)
1	1,522 ± 29	$806 \pm 32$	$749 \pm 33$	12 ± 2	$47 \pm 2$	$0.9 \pm 0.1$	$157 \pm 47$	13.8
2	$1,536 \pm 21$	$819\pm47$	$707 \pm 59$	$12 \pm 4$	$47 \pm 3$	$0.9 \pm 0.1$	$134 \pm 26$	14.0
3	$1,490 \pm 6$	$789 \pm 42$	$709 \pm 16$	$9 \pm 2$	$47 \pm 3$	$0.9 \pm 0.1$	$135 \pm 25$	13.5
4	$1,\!482\pm47$	$580 \pm 59$	$902 \pm 90$	$2 \pm 2$	$61 \pm 5$	$1.6 \pm 0.3$	$177 \pm 16$	9.9
5	$1{,}535 \pm 13$	$598 \pm 59$	$953\pm 64$	$4 \pm 2$	$61 \pm 4$	$1.6 \pm 0.2$	$198 \pm 25$	10.2

Average process performance observed during each experimental phase, under steady state conditions (Phase 6 is not considered, due to process instability)

Starting from day 70 (Phase 4), the increase in Alk/N molar ratio to 1.3 led to a corresponding increase in both NH<sub>4</sub>-N removal efficiency (from  $47\% \pm 3\%$  to  $61\% \pm 5\%$ ) and effluent NO<sub>2</sub>/NH<sub>4</sub> molar ratio (from  $0.9 \pm 0.1$  to  $1.6 \pm 0.3$ ), compared with previous phases. Decreasing DO concentration to 1.5 mg O<sub>2</sub>/L (Phase 5) did not cause any significant change in overall process performance, suggesting that limiting conditions did not occurred even below the DO concentration usually adopted in conventional WWTPs.

Table 2

However, the further decrease in DO concentration from 1.5 to 1.0 mg  $O_2/L$  (Phase 6) led to the irreversible worsening of process performance (Fig. 2): ammonia started to accumulate and pH raised up to 7.5 due to the lower alkalinity removal. After few days an almost complete washout of biomass was observed. Maximum FA concentration in the reactor was estimated around 50 mg NH<sub>3</sub>/L, much lower than those indicated as inhibiting by Van Hulle et al. [14]; therefore, the worsening of process performance was ascribed to the occurrence of oxygen limiting conditions. The threshold level observed in this study for DO concentration (1.5 mg  $O_2/L$ ) may depend on several factors (e.g., reactor configuration and low HRT), and is consistent with the broad range of DO threshold concentrations reported in previous studies: Guisasola et al. [16] indicated an oxygen affinity constant

for autotrophic ammonium oxidation within the range 0.16–2.0 mg O<sub>2</sub>/L; Van Hulle et al. [14] observed the worsening of process performance in a Sharon reactor operating at DO concentration lower than 3 mg O<sub>2</sub>/L; more recently, PN SBRs were successfully operated at DO concentration even lower than 1 mg O<sub>2</sub>/L [12,17]. Although it cannot be excluded that fine tuning of process parameters would allow to reduce the DO threshold concentration, such investigation was out of the scope of this study.

# 3.2. N<sub>2</sub>O emissions

As long as process performance was good and stable in terms of AOR and ammonium conversion to nitrite (Phases 1–5),  $N_2O$  emissions in the off-gas remained very low (average  $N_2O$  concentration in the off-gas and ER ranged between 2.5–3.2 ppmv and 2.1–2.7 mg N/L d, respectively), regardless of the applied DO concentration (Fig. 3(a)). Coherently, the increase in AOR due to the higher Alk/N molar ratio applied during Phases 4 and 5 was accompanied by a significant reduction of EF, from 0.35% (Phases 1–3) to 0.23% (Phases 4–5) of AOR (Fig. 3(b)). The low emission factors observed during Phases 1–5 can be likely ascribed to the positive effect of continuous mixing and



Fig. 3. Process performance in terms of AOR and  $N_2O-N$  emission rate (a), and  $N_2O-N$  emission factor (b), observed during the experimental campaign.

Reference	PN-reactor type	Influent type	Influent NH <sub>4</sub> -N (mg/L)	HRT (d)	NLR (g N/L d)	DO (mg/L)	N <sub>2</sub> O-N emission factor (per influent NH <sub>4</sub> -N)	N <sub>2</sub> O-N emission factor (per oxidized NH <sub>4</sub> -N)
This study <sup>a</sup>	Chemostat,	Synthetic	1,535 ± 13	1.0	1.5	1.5	0.14%	0.23%
	lab-scale							
[8]	SBR, lab-scale	Synthetic	300-1,000	1.0	0.3–1.0	0.5–1.0	-	0.11%-0.90%
[12]	SBR, lab-scale	Synthetic	600	0.5	1.2	0.35-0.85	0.57%-2.35%	-
[18]	Chemostat, full scale	Reject water	1,200–1,600	2.0-3.0	0.4-0.8	2.5	1.7%	3.4%
[20]	Air-lift continuous	Anaerob.	$1,\!500\pm0.19$	1.3–1.7	0.88-1.15	>2	0.6%-2.6%	-
	reactor, lab-scale	digested						
		black water						
[21]	SBR, lab-scale	Synthetic	1,000	1.0	8.0	0.5–0.8	-	1.0%
[22]	Granular SBR,	Synthetic	350	0.3	1.0	2.0	$0.8\%\pm0.4\%$	$1.5\%\pm0.8\%$
	lab-scale							
[23]	Granular airlift continuous reactor,	Reject water	$726 \pm 50$	0.4–0.6	0.85	4.4–6.7	-	$2.2\% \pm 0.4\%$
	pilot scale							
[23]	Granular airlift SBR,	Reject water	$450 \pm 78$	0.4–0.6	Approx.	5.7–7.2	-	19.3% ± 7.5%
	pilot scale				0.9			

Table 3	
Schematic comparison	among results reported in literature

<sup>a</sup>Values observed during Phase 5 (best performance).

aeration, which minimized the occurrence of anoxic conditions in the laboratory-scale reactor even at relatively low DO concentrations (1.5 mg  $O_2/L$ , Phase 5), thus reducing the occurrence of nitrifier denitrification. Indeed, such results look promising, compared with those previously reported in literature, although a direct comparison may be difficult, since the broad range of reported N<sub>2</sub>O ERs refer to very different system configurations (i.e., chemostat, continuous flow stirred tank reactor, sequencing batch reactor), size (i.e., laboratory-scale, pilot plant, full-scale plant) and operating conditions [2,18,19].

De Graaff et al. [20] treated the liquid effluent of a UASB reactor using a continuous flow reactor without biomass retention, with a DO concentration above 2 mg O<sub>2</sub>/L, and detected a EF ranging between 0.6% and 2.6% of total nitrogen load (0.14%–0.30% in this study); Law et al. [21] determined an average EF of  $1.0\% \pm 0.1\%$  of total ammonium converted in a PN-SBR; laboratory-scale PN-SBR systems, fed with synthetic influents and operated at different DO levels, were also studied by Rathnavake et al. [22] (DO = 2 ppm), who reported quite variable nitrous oxide EFs, averaged out at  $1.5\% \pm 0.8\%$  of the converted ammonium, and Kinh et al. [8], who observed a N<sub>2</sub>O EF of 0.11%-0.90% of oxidized ammonium, depending on pH, with DO kept in the range of 0.5–1.0 ppm: in both cases hydroxylamine oxidation, which is proportional to ammonia oxidation rate, was identified as the major N<sub>2</sub>O production contributor. On the other hand, a different behavior was observed in our study, since nitrous oxide EF decreased as AOR increased (Phases 4-5), thus indicating the minimization of hydroxylamine oxidation-driven N<sub>2</sub>O production at DO levels of 2 ppm or lower. Pijuan et al. [23] studied the effect of different DO concentrations on N<sub>2</sub>O emissions from a continuous pilot-scale granular airlift reactor performing both full and PN: the lowest EF

(2.2% of total converted ammonium) was measured at DO concentrations above 4.5 mg  $O_2/L$  (when DO was reduced, a proportional increase of  $N_2O$  EF was observed, up to 6% of converted ammonium); a similar behavior was reported by Lv et al. [12] who achieved stable PN in a SBR operating at oxygen-limiting conditions (DO = 0.35–0.85 mg  $O_2/L$ ), and measured  $N_2O$  emissions ranging from 0.57% to 2.35% of total influent nitrogen; conversely, in this study the EF decreased as DO decreased (Phases 1–5), as long as limiting conditions did not occur. A schematic comparison among results reported in literature and those achieved in this study is shown in Table 3.

As DO concentration was further reduced to 1 mg O<sub>2</sub>/L (Phase 6), the sudden increase in both N<sub>2</sub>O ER (>4.5 mg N/L d) and EF (up to 0.61% of AOR) was observed, consistently with the worsening of the overall process performance described previously. At DO concentrations below 1.5 mg O<sub>2</sub>/L, the shortage of available oxygen likely caused the increase in the anoxic formation of N<sub>2</sub>O due to nitrifier denitrification, which has been recognized as the main pathway contributing to N<sub>2</sub>O production [3]. Law et al. [24] observed an opposite behavior (i.e., the decrease in N<sub>2</sub>O production with decreasing DO concentration) in AOB cultures previously adapted to low DO concentrations (0.5-0.8 mg O<sub>2</sub>/L) and exposed to relatively high NH<sub>4</sub>-N and NO<sub>2</sub>-N concentrations (500 mg N/L); however, influent NH<sub>4</sub>-N concentration was much higher in our study (1,500 mg N/L), so that complete AOB acclimation to low DO levels may not be enough to achieve stable PN.

# 4. Conclusions

In this study, a PN reactor was fed with a synthetic medium at a constant NLR of 1.5 g N/L d, and N<sub>2</sub>O gaseous

emissions were measured with different applied Alk/N molar ratios (1.0-1.3) and DO concentrations (5.0-1.0 mg  $O_2/L$ ). As DO concentration was not limiting, stable process performance was achieved in terms of AOR and ammonium conversion to nitrite, and N<sub>2</sub>O emissions in the off-gas were lower than most of the values reported in previous studies. The increase in influent Alk/N was accompanied by the corresponding increase in AOR, while N<sub>2</sub>O ERs did not change significantly, thus resulting in the reduction of the N<sub>2</sub>O-N EF. Unlike many of the results previously reported in literature, where increase in AOR or decrease in DO level appeared to trigger nitrous oxide production, reactor configuration adopted in this study (i.e., nonaerated settling and discharge phases were avoided) coupled with a continuous aeration strategy led to minimization of anoxic conditions. As a consequence, this contributed to the reduction of N<sub>2</sub>O emissions even at low, as long as not process-limiting, DO concentrations. As DO was set at 1.0 mg O<sub>2</sub>/L, overall process performance was irreversibly compromised: beside the drop in AOR, a corresponding increase in N<sub>2</sub>O-N EF was observed. Nitrifier denitrification, more than hydroxylamine oxidation, was suggested as the main pathway contributing to N<sub>2</sub>O formation.

Results showed that the environmental footprint of PN process can potentially be reduced by applying proper aeration strategy at relatively low DO concentrations; moreover, chemostat reactor configuration can represent a suitable choice even at high N load. Results are promising, and further investigation in this sense will be carried out treating real wastewater.

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# Symbols

- *c* N<sub>2</sub>O level in the gas sample, ppmv
- $ER N_0O-N$  emission rate, mg N/L d
- $M_{\rm M}$  Molar mass of nitrogen, 14.0067 g/mol
- *p* Atmospheric pressure, 1 atm
- Q Volumetric flow rate of the off-gas, L/d
- R Gas constant, 0.082056 L atm/mol K
- *T* Temperature, K
- $V_{I}$  Working volume of the reactor, L

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