

## Partial Nitrification of Wastewater from an Aminoplastic Resin Producing Factory

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### Abstract

Nitrification via nitrite was studied in two aerobic reactors treating wastewater from an aminoplastic resin producing factory at HRT varying between 1.37-1.89 and 2.45-3.63 days. Both reactors were fed with concentrations of 366, 450, 1099 and 1899 mg N-NH<sub>4</sub><sup>+</sup>/L. In general in the reactor operated at a lower HRT, the nitrification percentage decreased from 87.2 to 21.6%, while the nitrification percentage remained always lower than 2.5% (except in the last period) when the ammonium concentration was increased. This behaviour could be due to the inhibition of the ammonium and nitrite oxidation produced by high free ammonia concentrations up to 179.3 mg N-NH<sub>3</sub>/L. In the reactor operated at a higher HRT, the nitrification percentage decreased and the nitrification percentage increased from 88.6 to 39.6% and from 0.65 to 35.7%, respectively, due to an increase of the dissolved oxygen concentration from 0.76 to 1.02 mg O<sub>2</sub>/L. However, when ammonium was fed at a concentration of 1898.7 mg N-NH<sub>4</sub><sup>+</sup>/L, the nitrification increased and the nitrification decreased, probably as a result of the accumulation of free ammonia up to 2.04 mg N-NH<sub>3</sub>/L, meaning that nitrite oxidizers were inhibited. Nitrite build-up was observed after each modification of ammonium concentration in the feed.

### Keywords

Industrial wastewater; nitrification; via nitrite

### Notation

i	influent
e	effluent
DO	dissolved oxygen
FA	free ammonia FNA free nitrous acid
HRT	hydraulic retention time
VSS	volatile suspended solids

## Introduction

Nitrification and denitrification are well known processes used for treating nitrogen containing wastewater. Recently, a better effluent quality demand as well as the need of more cost-effective wastewater treatment processes have induced the research for new biological wastewater treatment systems. Although it is known that nitrite is a toxic compound to the aquatic organisms; nitrification and denitrification could be improved if the nitrification is stopped at the level of the intermediate nitrite. This could lead to the following potential cost savings (Turk and Mavinic, 1987):

- the oxygen requirements during the aerobic phase are 25% lower
- carbon requirements for denitrification are 40% lower
- the nitrite reduction rate is approximately double than nitrate reduction rate.
- in anoxic phase less biomass is produced.

Nitrite oxidation rate is generally faster than the ammonium oxidation rate so that nitrite rarely accumulates in the medium. But several factors such as free ammonia, free nitrous acid, low dissolved oxygen concentration, pH, temperature, could selectively inhibit nitrite oxidisers making possible nitrite build-up.

Both FA and FNA (non-ionized forms of ammonium and nitrite) are well-known nitrification activity inhibitors, being Nitrobacter more sensitive to the effects of these compounds. This nitrification inhibition results in nitrite accumulation. FA and FNA concentrations in the system depend on pH and T and can be calculated by means of the following equations:

$$[N - NH_3] (mg/L) = \frac{[N - NH_4^+]_{reactor} (mg/L) * 10^{pH}}{K_b/K_w + 10^{pH}}$$

$K_b$  = ionization constant of ammonia in the equilibrium equation.

$K_w$  = ionization constant of water.

$$K_b/K_w = e^{(6.344/273+7)}$$

$$[N - HNO_2] (mg/L) = \frac{[N - NO_2]_{reactor} (mg/L)}{K_a * 10^{pH}}$$

where  $K_a$  is an ionization constant of  $HNO_2$  equilibrium and can be determined by the following expression:

$$K_a = e^{(-2.300/273+T)}$$

As mentioned, FA and FNA concentrations depend on pH, so that at high pH a large part of the ammonium will be present as ammonia and at low pH most of the nitrite will be present in the form nitrous acid.

The concentrations of FA and FNA inhibitors were estimated by many authors. Anthonisen et al. (1976) found FA inhibition of Nitrobacter and Nitrosomonas in the ranges in 0.1 – 1.0 mg  $NH_3/L$  and 10 – 150 mg  $NH_3/L$  respectively and FNA inhibition of nitrifying organisms at 0.22 – 2.88 mg  $HNO_2/L$ . This point was corroborated by Liu and Capdeville (1994). Nevertheless, other authors suggest higher threshold inhibitors concentrations. Acclimation of biomass to high ammonia concentrations is thought to be one of the factors behind this discrepancy (Turk and Mavinic, 1989).

The aim of the present work was to study the feasibility of achieving nitrification via nitrite using sludge from the aerobic chamber of an industrial wastewater treatment

plant which was operated under overall nitrification conditions. For this purpose the dissolved oxygen was controlled and maintained at a low concentration and the free ammonia and free nitrous acid concentrations were frequently checked in the reactors.

## Material and methods

### *Wastewater and reactors*

Sludge from the aerobic chamber of a wastewater treatment plant of an amino plastic resin producing industry was inoculated in two similar laboratory scale aerobic reactors operated at HRT varying between 1.37–1.89 and 2.45–3.63 days respectively. Both reactors were fed with the same substrate. The reactor consisted in a glass vessel with a useful volume of 1.5 litre connected to a micro DCU 300 system and a settler. DO on the vessel was controlled automatically by a pO<sub>2</sub> controller which activates the stirring, so that no pump was needed. Peristaltic pumps were employed for influent feeding and recirculation. The liquor mixture from the reactor is discharged into a settler in which sludge and supernatant are separated. Sludge is returned periodically into the reactor and maintained in the system with a minimum purge to the concentrations of solids measures and for analytical reasons.

The reactors were kept in a thermostatic chamber at 20 °C, but the averaged operating temperature in the reactor was 23.1 °C.

The aerobic reactors were fed with the effluent of a laboratory scale anoxic reactor treating wastewater from an amino plastic resin producing factory. In the anoxic reactor the organic compounds were oxidized to CO<sub>2</sub>, and organic nitrogen was hydrolyzed to ammonium.

The effluent of the anoxic reactor is filtered and the ammonium concentration is adjusted by adding NH<sub>4</sub>Cl and (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>. Since the alkalinity of the wastewater was not high enough to develop the nitrification, additional alkalinity was added to maintain a NaHCO<sub>3</sub>/N-NH<sub>4</sub><sup>+</sup> ratio of 10. Micronutrients (Eiroa et al., 2004) were added with a dilution 1:2000.

### *Analytical methods*

Ammonium, nitrite and nitrate and pH in the effluent were determined three times per week. Once a week VSS were measured in the reactor and effluent. Ammonium and nitrite were analysed in the reactor once a week in order to obtain exposure levels of the biomass to FA and FNA.

Except samples collected to measure ammonium and pH, the rest of the samples were filtered through nylon syringe filters (0.45mm) to separate the liquid and solid phases preventing further reactions. Nitrite and nitrate anions were analysed by Capillary Electrophoresis (Hewlett Packard) with a micro capillary tube of fused silica. A sodium phosphate solution was employed as the electrolyte and UV detection was undertaken at a wavelength of 214 nm and 450 nm as reference. Ammonium and DO concentrations were measured with a selective electrode. VSS and pH were evaluated according to Standard Methods (APHA, 1998).

### *Calculations*

$$\text{Ammonium oxidation: } 100 * \frac{[N-NO_2^-]e-[N-NO_2^-]i+[N-NO_3^-]e-[N-NO_3^-]i}{[N-NH_4^+]i}$$

$$\text{Nitrite accumulation: } 100 * \frac{[N-NO_2^-]e-[N-NO_2^-]i}{[N-NO_2^-]e-[N-NO_2^-]i+[N-NO_3^-]e-[N-NO_3^-]i}$$

$$\text{Nitritation percentage: } 100 * \frac{[N-NO_2^-]e - [N-NO_2^-]i}{[N-NH_4^+]i}$$

$$\text{Nitrataion percentage: } 100 * \frac{[N-NO_3^-]e - [N-NO_3^-]i}{[N-NH_4^+]i}$$

## Results and discussion

During the experimental period different nitrogen loading rates (NLR) were applied. For reactor R1, operated at a HRT close to 1.7 d, the nitrogen loading rate ranged from 0.254 to 1.164 g N-NH<sub>4</sub><sup>+</sup>/L·d and for reactor R2, operated at a HRT close to 3.0 d, the nitrogen loading rate ranged from 0.108 to 0.645 g N-NH<sub>4</sub><sup>+</sup>/L·d.

Table 1 summarises the operational conditions maintained in reactors R1 and R2 during the experimental period. DO was checked and maintained close to 0.8 mg O<sub>2</sub>/L. Previous assays showed this concentration was the optimum level to achieve nitrification via nitrite. To try to enhance ammonium oxidation in R1, DO concentration was increased to 2.54 mg O<sub>2</sub>/L in the last period (E). In R2, DO level was decreased to 0.43 mg O<sub>2</sub>/L in the period E to avoid nitrite oxidation to nitrate.

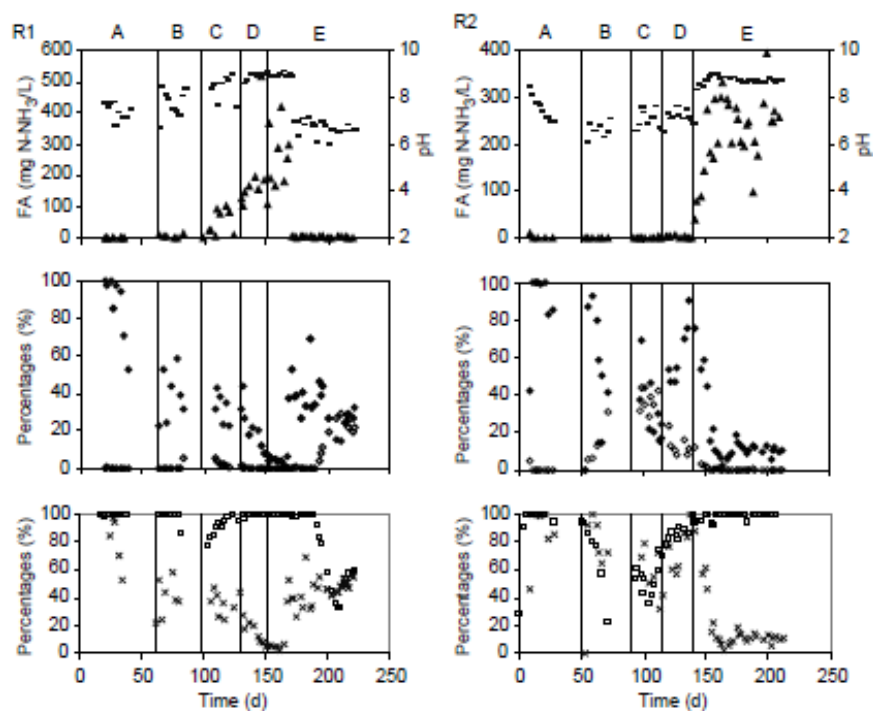
Table 1 Operational conditions of reactors R1 and R2

		N-NH <sub>4</sub> <sup>+</sup> (mg/L)				
		(A) 450	(B) 366	(C) 1099	(D) 1899	(E) 1899
R1	Operation day	17-39	63-83	103-122	123-152	153-222
	NLR (kg/m <sup>3</sup> ·d)	0.336	0.254	0.753	1.0806	1.164
	DO <sub>r</sub> (mg O <sub>2</sub> /L)	0.82	0.83	0.65	0.63	2.54
R2	Operation day	6-28	52-72	92-113	114-140	141-212
	HRT (d)	3.63	3.18	2.45	2.97	3.31
	NLR (kg/m <sup>3</sup> ·d)	0.126	0.108	0.504	0.645	0.631
	DO <sub>r</sub> (mgO <sub>2</sub> /L)	0.76	0.89	1.02	0.99	0.43

### Time course analysis

Both reactors were fed with concentrations of 366, 450, 1099 and 1899 mg N-NH<sub>4</sub><sup>+</sup>/L.

In R1, during periods A-D the DO concentration was kept between 0.63 and 0.83 mg O<sub>2</sub>/L and the ammonium concentration in the feed was varied between 366 and 1899 mg N-NH<sub>4</sub><sup>+</sup>/L. Ammonium oxidation decreased from 90.2 to 20.0% with a simultaneous FA accumulation from 0.15 to 179.34 mg N-NH<sub>3</sub>/L. The nitritation percentage decreased from 87.2 to 21.6%, while the nitrataion percentage remained always below 2.9%. The nitrite accumulation was always higher than 76.9%. As shown in Figure 1, in period A an ammonium oxidation of 90.2% and a nitrite accumulation of 99.8% were obtained with a FA concentration of 0.15 mg N-NH<sub>3</sub>/L, which was not inhibitory to ammonium oxidizers but it was inhibitory to nitrite oxidizers. However, in period B the FA concentration reached an average of 5.8 mg N-NH<sub>3</sub>/L which was inhibitory to both ammonium and nitrite oxidizers, resulting in an ammonium oxidation of 39.7% and in a nitrite accumulation of 97.9%.



**Figure 1** Evolution of free ammonia ( $\blacktriangle$ ) and pH ( $\blacksquare$ ); nitritation ( $\blacklozenge$ ) and nitrification ( $\diamond$ ) percentages; and ammonium oxidation ( $\times$ ) and nitrite accumulation ( $\square$ ) in R1 and R2. The ammonium concentration in the feed at the different periods was: 450 (A), 366 (B), 1099 (C), 1899 (D) and 1899 (E) mg N-NH<sub>4</sub><sup>+</sup>/L

In period C the average ammonia concentration increased to 58 mg N-NH<sub>3</sub>/L, being the ammonium oxidation of 35.3%, close to that obtained in period B. During period D the FA concentration rose from 125 to 187 mg N-NH<sub>3</sub>/L, and an almost complete inhibition of ammonium oxidizers was observed, reaching values of ammonium oxidation close to 8%. In both periods C and D, the DO concentration was similar around 0.64 mg O<sub>2</sub>/L. However, this DO concentration was not a limiting factor since the nitrite concentration in period D averaged 399 mg N-NO<sub>2</sub>/L with a maximum close to 680 mg N-NO<sub>2</sub>/L and higher than in period C (302 mg N-NO<sub>2</sub>/L). The increase of ammonium concentration in the feed together with excess of alkalinity caused an increase of pH and FA resulting in a higher inhibition of nitritation. In period E, the ammonium concentration in the feed was the same as in period D (1899 mg N-NH<sub>4</sub><sup>+</sup>/L). In order to improve the ammonium oxidation DO concentration was increased from 0.64 to 2.54 mg O<sub>2</sub>/L. At the beginning of this period, the alkalinity in the feed was 13170 mg CaCO<sub>3</sub>/L and the reactor pH was close to 8.7 with an average FA concentration increase of 257.7 mg N-NH<sub>3</sub>/L being the ammonium oxidation close to 9.3%. Hence, the alkalinity was decreased in the feed down to a NaHCO<sub>3</sub>/N-NH<sub>4</sub><sup>+</sup> ratio equal to 6, leading to a decrease of the pH to 6.77 and a decrease of FA concentration to 3.9 mg N-NH<sub>3</sub>/L. Under these conditions ammonium oxidizers recovered their activity, and the nitritation percentage increased to 41.2%, with nitrite accumulation of 98.8%. However at the end of the period an increase in nitrite oxidation took place along with a decrease of FA concentrations with a final concentration of 3.0 mg N-NH<sub>3</sub>/L. In R2 during periods A, B and C, the DO concentration increased from 0.76 to 1.02 mg O<sub>2</sub>/L and the nitrite accumulation decreased from 90.9 to 44%. So, nitritation decreased from 88.6 to 39.6% while nitrification increased from 0.6 to 35.6%. According to Ruiz et al. (2003) this behaviour can be due to the DO concentration. They obtained a maximum nitrite accumulation of 65% at 0.7 mg O<sub>2</sub>/L, but at higher DO concentrations nitrite was oxidized to nitrate. In

addition, the FA concentration decreased from 1.72 to less than 0.4 mg N-NH<sub>3</sub>/L. With regard to ammonium oxidation, in spite of remaining higher than 73.3%, a slight decrease was observed simultaneous to an increase of the nitrogen loading rate from 0.108 to 0.504 g N-NH<sub>4</sub><sup>+</sup>/L·d. This tendency was also found by Gupta and Sharma (1996) who observed that the nitrogen removal efficiency decreased when the nitrogen loading rate increased. In period D, the DO concentration was close to the DO concentration of period C (1.02 mg O<sub>2</sub>/L) and the nitrogen loading rate was only increased 1.25 times (this varied between 0.504–0.645 Kg/m<sup>3</sup>·d). Under these conditions ammonium oxidation was approximately the same, but nitrification percentage raised to 50.8% and nitratation percentage went down to 16.7%. Possibly this could be related to an increase in the FA concentration to 2.04 mg N-NH<sub>3</sub>/L, meaning that nitrite oxidizers were inhibited by free ammonia. However, ammonium oxidizers were not inhibited by this free ammonia concentration. In period E, in order to achieve a higher nitrification efficiency a DO concentration of 0.43 mg O<sub>2</sub>/L was tested, but the oxygen became a limiting factor. Consequently the ammonium oxidation decreased to 18.4%.

In most of the periods and for both of the reactors nitrite build-up was observed after each modification of the ammonium concentration in the feed, probably associated to a variation of the rest of the parameters involved in this process, as pH, DO... High nitrite concentrations were reached in both reactors, being the highest nitrite concentrations 1597 mg N-NO<sub>2</sub>/L in R2 (period D) and 1194 mg N-NO<sub>2</sub>/L in R1 (period E).

Nitratation percentages were very different in both reactors. The highest nitrate percentages were obtained in R2 while highest nitrite accumulations were attained in R1. This difference can be due not only to the free ammonia and DO found in the medium but also to the HRT used. Lower nitratation percentages obtained for lower HRT which is probably associated to the lower contact time between bacteria and nitrogen substrate. Ruiz et al. (2003) working with a SBR reported that at short aerobic phases nitrite accumulation was greater than at long aerobic phases, however, nitrogen removal was more efficient in long aerobic phases. The DO and the medium free ammonia are important parameters for nitrite build-up.

### *DO influence*

DO is known to have an important effect on the activity and settleability of the sludge (Dangcong et al., 2001). Generally, in overall nitrification process it is recommended to keep DO levels higher than 2–3mg O<sub>2</sub>/L to obtain nitrate as final product. Several authors observed that working with lower levels they were able to stop the process at the intermediate nitrite (Ruiz et al., 2003; Garrido et al., 1997).

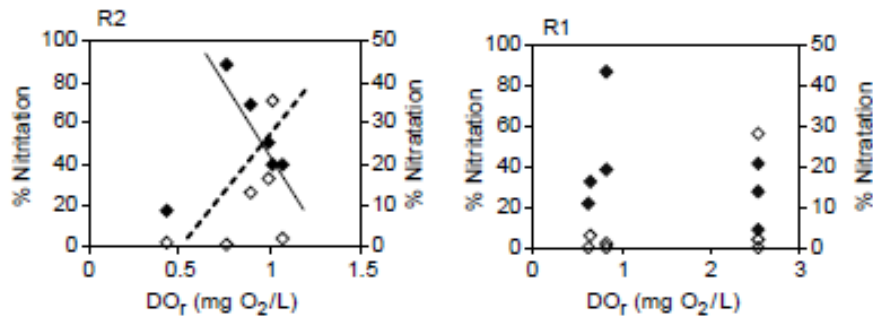
As shown in Table 1, the DO concentration in R1 decreased during the course of the experimental period except in the last assay where it was increased to 2.54 mg O<sub>2</sub>/L.

As can be seen in Figure 2, in R2, the DO concentration ranged from 0.76 to 1.02 mg O<sub>2</sub>/L being in the last period decreased to 0.43 mg O<sub>2</sub>/L. A nitrification percentage increase was noticed with a DO concentration decrease up to reach a value of 0.76 mg O<sub>2</sub>/L, at which a maximum nitrification percentage of 88.6% was attained. At DO concentration of 0.43 mg O<sub>2</sub>/L nitrification and ammonium oxidation fell down to 17.6% and 18.4% respectively. This indicates that the DO concentration is a limiting factor in nitrification, which was also reported by Yoo et al. (1999) who observed a decrease or suppression of nitrification at low DO concentrations. At this low DO concentration a decrease of ammonium oxidation and alkalinity consumption resulting in a FA

concentration increase. The FA concentration reached in this period was high enough to inhibit both the nitritation and nitrification processes. The nitrification percentage increased from 0.6 to 35.6% with the DO, as can be seen in Figure 2.

The results obtained in this research are in agreement with the results obtained by Ruiz et al. (2003) who observed an increased nitrite build-up and the same ammonium consumption at DO concentrations ranging between 1.7–0.7 mg O<sub>2</sub>/L. Both nitrite buildup and ammonia oxidation decrease appeared when the DO concentration was below 0.5 mg O<sub>2</sub>/L. Nitrite build-up started at concentrations below 1.7 mg O<sub>2</sub>/L, reaching a maximum nitrite build-up at 0.7 mg O<sub>2</sub>/L, being ammonium completely oxidized.

In R1 a nitritation percentage higher than 21.6% was obtained at values of DO from 0.63 to 2.54 mg O<sub>2</sub>/L. A maximum nitritation percentage of 87.2% and ammonium oxidation of 90.2% were achieved at 0.82 mg O<sub>2</sub>/L. Nitritation percentages of 32.4% and 21.6% were attained at DO concentrations of 0.65 and 0.63 mg O<sub>2</sub>/L respectively.



**Figure 2** Nitritation (◆) and nitrification (◇) percentages of R1 and R2 versus average DO levels of different operational periods for reactors R2 and R1

However, at DO concentrations of 0.83 mg O<sub>2</sub>/L a nitritation percentage of 38.9% was also observed, corresponding to a different operation period of the reactor. At a DO concentration of 2.54 mg O<sub>2</sub>/L, wide variations of nitritation (9.3–41.2%) and nitrification percentages (0–20.9%) were obtained, probably due to variations in FA concentration (258–2.3 mg N-NH<sub>3</sub>/L). In both periods D and E the nitritation percentages obtained were always lower than at a DO concentration of 0.82 mg O<sub>2</sub>/L, which may indicate that other inhibitory factors must be considered. The nitrification percentage was maintained below 2.9% except for a DO concentration of 2.54 mg O<sub>2</sub>/L in which the nitrification percentage increased from 0 to 20.9%. With regard to ammonium oxidation, the tendency expected would be to obtain low ammonium oxidation at concentration below 0.82 mg O<sub>2</sub>/L and higher oxidation at higher concentrations than 0.82 mg O<sub>2</sub>/L. However, in this work at DO concentrations higher than 0.82 mg O<sub>2</sub>/L we observed a decrease of ammonium oxidation, which could indicate that DO is not sole factor to take into account.

Analysing periods (D) and (E) in which ammonium concentration in the feed was close to 1899 mg N-NH<sub>4</sub>/L and DO concentration was modified, it was observed that in R1 when the DO concentration was increased from 0.63 to 2.54 mg O<sub>2</sub>/L, the percentage ammonium oxidation increased from 20.0 to 49.0% but nitrite accumulation decreased from 99.3 to 53.7%. In R2 when DO concentration was decreased from 0.99 to 0.43 mg O<sub>2</sub>/L ammonium oxidation dropped from 67.4 to 18.4%, but nitrite accumulation

rose from 73.8 to 98.4%. Hence, limiting DO concentration was the main factor in order to attain nitrification via nitrite.

### FA and FNA effects

As shown in Figure 3, in both reactors a FA concentration increase caused a nitritation percentage decrease. With regard to nitrataion percentage, in R1 for periods A-D it was lower than 2.9% at FA concentrations ranging from 0.15 to 179.34 mg N-NH<sub>3</sub>/L. At the beginning of period E a nitritation percentage of 5.9% and nitrataion percentage of 0% were obtained when the average FA concentration in the reactor was 258 mg N-NH<sub>3</sub>/L. At the end of the period E the FA concentration decrease to 2.3 mg N-NH<sub>3</sub>/L leading to an increase of nitritation percentage to 28%. Due to the low FA concentration and high DO concentration, a nitrataion percentage of 20.9% was obtained. It is believed that DO alleviate FA inhibitor effect, so discrepancies in FA inhibitors concentrations reported by different authors might be due to this fact (Turk and Mavinic, 1989). On the other hand, Suthers and Ganczarczyk (1986) working at DO concentrations between 6-8 mg O<sub>2</sub>/L noticed that *Nitrobacter* before continuously acclimatizing was able to tolerate inhibitory concentrations of FA equal to 2.5 mg N-NH<sub>3</sub>/L at which it was previously inhibited. In R2 the FA concentrations in periods B, C and D were lower than those found in R1, obtaining higher nitrataion percentages than in R1. In both reactors FNA inhibition of nitrifying organisms was not observed in spite of reaching FNA concentrations in the range proposed as inhibitory by Anthonisen et al. (1976).

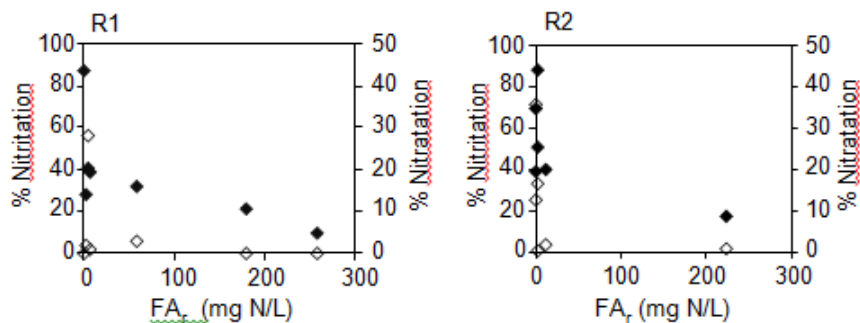


Figure 3 Nitritation (♦) and nitrataion (◇) percentages of R1 and R2 versus median FA levels of different operational periods

### Conclusions

Differences in DO concentration, FA concentration, nitrogen loading rate and HRT could explain different nitritation, nitrataion and ammonium oxidation percentages obtained in this research.

An optimum DO concentration of 0.8 mg O<sub>2</sub>/L led to the maximum nitritation percentage, higher DO concentrations might cause a decrease in nitritation and an increase in nitrataion percentages. A deviation in this tendency might indicate that other factors such as an increased nitrogen loading rate and FA might be influencing the nitrification.

A FA concentration increase provoked a nitritation percentage decrease. Different nitrataion percentages obtained for R1 and R2 could be explained with the different FA



concentration present. FA concentrations higher than 2.04 mg N-NH<sub>3</sub>/L were inhibitory.

At high HRT the reactor showed higher nitrataion percentages probably associated to the higher contact time between bacteria and substrate.

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