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Denitrification via Nitrite in a Modified UASB reactor using Chilean zeolite as Microbial Support

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ABSTRACT: The general objective of this study was to develop a highly efficient, economical and integrated technology for the removal of nitrogen compounds through denitrification via nitrite. To achieve this, a modified UASB reactor was designed, set-up and operated using Chilean zeolite as microbial support. The results were compared to a conventional UASB reactor used as control. The volume of each reactor was 2 L. The reactors operated with synthetic wastewater under the same operating conditions (with superficial velocities, v_s, of up to 1 m/h) in the first part of the experiment. Later, during the second part of the experiment, only the modified UASB was used, with v_s of up to 5.5 m/h. In the first part of the experiment, a higher velocity of denitrification in the reactor with zeolite was obtained. Nitrogen removal at the end of this experimental set for both reactors, with a v_s lower than 1 m/h, was 87%. In the second stage, the modified UASB reactor operated at v_s of between 1.5 and 5.5 m/h. Here, it was observed that the removal of nitrite increased significantly. Specifically, at v_s values of 2.5, 4.0 and 5.5 m/h, a value of the nitrogen loading rate (NLR) of 1.22 kg $N\text{-}NO_2^-/m^3/d$ was kept constant, achieving nitrogen removal efficiencies of 50%, 65% and 95.5% respectively. This last value proves how highly effective the modified UASB reactor is when operating with v_s as high as 5.5 m/h.

Key words: Denitrification via nitrite, Modified UASB, Zeolite, Microbial support

INTRODUCTION

Biological treatments for the elimination of nitrogen from wastewaters are the most frequently used processes. They are more advantageous in terms of cost than physical-chemical processes. The application of this type of treatments is generally carried out with the aim of decreasing the concentration of organic compounds and nitrogen. Nitrogen is found in its different forms in wastewaters (Chen *et al.*, 2000; Ferzani *et al.*, 2005; Hill *et al.*, 2005; Mtelhiwa *et al.*, 2008; Taseli, 2009; Coetzee *et al.*, 2011). It is a by-product of industrial processes mainly from the fertilizer, food, agricultural and livestock industries and can frequently cause the contamination of the course of water receptors (Montalvo *et al.*, 2011). Furthermore, these biological treatments entail certain disadvantages. For example, in total nitrification, the nitrification of water with high nitrogen loads implies elevated aeration costs. In denitrification, an adequate C/N ratio may not be achieved, due to the fact that the previously mentioned processes, which are commonly applied, are carried out with the sole purpose of reducing organic matter.

Denitrification can be optimized through the nitrite route, which involves partial nitrification to obtain nitrite as well as a denitrification of this nitrite - instead of nitrate - obtaining considerable reductions in hydraulic retention times and a saving in operational costs (Azimi and Zamanzadeh, 2004; Ruiz *et al.*, 2006; Liu *et al.*, 2010). One of the biological reactors most commonly used in wastewater treatment is the UASB reactor, which is characterized mainly by its simplicity

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in construction, design and operation and its capacity to treat waters with high organic and nitrogen loads (Seghezzo *et al.*, 1998; An *et al.*, 2008; Wu *et al.*, 2011; Akbarpour and Mehrdadi, 2011; Arshad *et al*., 2011). One of the operational disadvantages of the UASB reactor is its low resistance to superficial velocities (v_s) higher than 1 m/h, which generates the washing of sludge from the interior of the reactor. In order to work at v_s higher than the previously mentioned value, a modification to the UASB was implemented, using a low cost support medium, such as zeolite, which would lead to a system with granular adhered biomass. This mineral possesses physical properties that provide a great adsorption capacity which facilitates the adherence of the biomass to the walls, and at the same time, resisting higher v_s, and, therefore, higher nitrogen loads (Fernández *et al.*, 2007; Andrade *et al*., 2008; Weiβ *et al.*, 2011).

MATERIALS & METHODS

During these experiments a comparison was made between a fixed biomass modified UASB and a suspended biomass conventional UASB which acted as the control throughout the study. The two reactors were built under the same conditions with the same volume of 2 L. A schematic diagram of both reactors is shown in Fig. 1, illustrating both the UASB and the modified UASB (named as such because it contains zeolite as a microbial support). Prior to the start-up of the reactors, their components were quantitatively determined. Once inoculated, the anaerobic biomass concentration in the reactors was 25 g VSS/L. This inoculum came from a UASB reactor operating at real scale. The inoculum had 57 g/L total suspended solids (TSS), 46.7 g/L volatile suspended solids (VSS) and a pH of 7.8.

The volume of zeolite (2 mm in diameter) used in the reactor was equivalent to 12% of the effective reactor volume. Table 1 shows the main characteristics of the zeolite used in the modified UASB reactor. A synthetic solution containing all the components listed in Table 2 was used as wastewater. From the values presented in this table, it can be seen that the C/N ratio

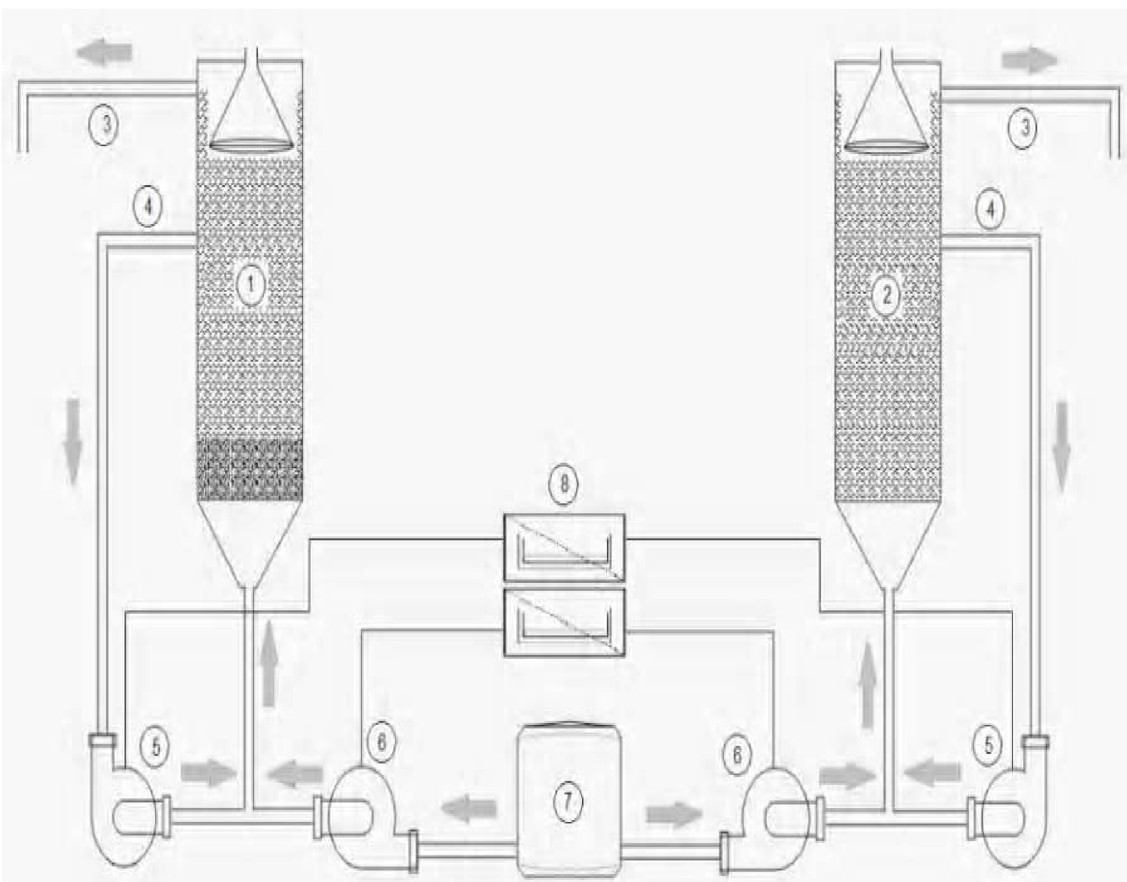

Fig. 1. Schematic diagram of the setup of the UASB reactors

1) Reactor with zeolite ; 2) Reactor without zeolite; 3) Effluent; 4) Recirculation; 5) Recycling pump; 6) Feeding pump; 7) Feed tank; 8) Timers

was kept constant at 4:1. Table 3 summarizes the working and operational conditions in each one of the stages considered in this process. These conditions were valid for both reactors as far as the fourth stage (experiment part I). The change to the following phase depended on the behavior of the nitrogen compounds and the level of organic matter degradation. Stages 5 to 8 (experimental study II) were only performed for the modified UASB reactor. Stage 1 corresponded to the start-up of the reactors; there was no feed and the reactors were operated in batch mode with total recirculation until a good microbial growth rate was achieved. In the case of the modified UASB reactor, continuous feeding started when maximum adherence and maximum nitrogen and organic matter degradation were achieved. The analysis of solids and nitrite were carried out according to Standard Methods for the Examination of Waters and Wastewaters (APHA, 2005). Nitrate, ammonium nitrogen and pH were determined by selective electrodes.

RESULTS & DISCUSSION

In the first part of the experiment, the results were obtained from the performance of both reactors, which were operated under the same conditions (stages 1 to 4). For stages 5 to 8 (called experiment part II), only the modified UASB reactor was operated due to the fact that the v_s values were higher than 1 m/h. During the start-up, total recirculation in the UASB reactors was performed. In the case of the modified UASB reactor

Table 1. Chemical and mineralogical composition of the Chilean zeolite used in the modified UASB reactor

Chemical composition (%)		Mineralogical composition (%)	
SiO_2	66.62	Clinoptilolite	35
Al_2O_3	12.17	Mordenite	15
Fe_2O_3	2.08	Montmorillolite	30
CaO	3.19	Others	20
MgO	0.77		
Na_2O	1.53		
K_2O	1.20		
Ignition Waste	11.02		

Table 2. Composition of the synthetic wastewater used as feed in the UASB reactors

Macronutrient Solution			Micronutrient Solution		
Compound	Unit	Quantity	Compound	Unit	Quantity
CH_3COOH	g/L	58	EDTA	g/L	0.15
$NaNO_2$	g/L	14	HCl	mL/L	1
Yeast Extract	g/L	2	$FeSO_4$	g/L	2
Na_2CO_3	g/L	10	HBr	g/L	0.05
K_2HPO_4	g/L	31.6	$ZnCl_2$	g/L	0.05
KH_2PO_4	g/L	25	$MgCl_2$	g/L	0.05
Micronutrients	mL/L	13.5			

Table 3. Operating parameters in each stage of the UASB reactor and modified UASB reactor

Stage	NLR (kg N-NO_2^-/m^3/d)	OLR (kg COD/m^3/d)	Q (mL/d) (feed)	Q (mL/d) (recycle)	COD (g O_2/L)	v_s (m/h)
1	-	-	0	35	58.4	0.13
2	0.16	1	35	215	58.4	0.94
3	0.48	3	130	120	46.2	0.94
4	0.81	5	290	0	34.8	1.09
5	0.81	5	400	0	25.2	1.5
6	1.22	7.6	650	0	23.6	2.5
7	1.22	7.6	1050	0	14.6	4.0
8	1.22	7.6	1450	0	10.6	5.5

with zeolite, this was not fed until a decrease in the concentration of nitrite was observed at which point the operation changed to continuous mode at a low NLR (1 kg N-NO_2^-/m^3/d), which was increased when the concentration of nitrite observed in the effluent was constant. The start-up phase, or stage 1, lasted 21 days. Fig. 2 shows that the average VSS concentrations found in the effluent of both UASB reactors were similar. The two values were of the same magnitude, although in the case where zeolite was not used the value was slightly higher. Where the reactor with zeolite averaged an effluent VSS concentration of 3.36 g/L, the reactor without zeolite had a concentration of 3.68 g/L. This indicates that at a v_s lower than 1 m/h, the presence of a support influences the concentration of VSS in the effluents from the reactors only to a small extent, giving somewhat lower concentrations in the reactor containing zeolite.

The effluent VSS concentrations in the modified UASB reactor corresponding to stages 5 to 8, where the reactor operated at a v_s of between 1.5 and 5.5 m/h, are summarized in Table 4. It can be observed that when the v_s increased, the effluent VSS concentration also increased, but still only slightly, e.g. when the v_s increased by 3.7, the effluent VSS content only rose by a factor of 1.2. This indicated how well this modified reactor performed. At a v_s of 5.5 m/h (stage 8) the hydraulic performance of the reactor behaved in a similar way to that of an expanded bed, since the sludge permanently occupied between 40% and 50% of the reactor volume. Granular sludge was also demonstrated to be very effective in a sequencing batch airlift reactor operating at a reduced aeration rate (Wang *et al.*, 2009). However, the OLR maintained in this reactor to obtain an effective operational performance (2.8 kg COD/m^3/d) was much lower than that achieved in the present modified UASB reactor (5.0-7.6 kg COD/m^3/d) operating during stages 5-8. In addition, OLR (2.7 kg COD/m^3/d) and NLR values (0.43 kg N-NO_2^-/m^3/d) much lower than those studied in the present work were used in a lab-scale sequencing batch reactor (SBR) seeded with granular sludge treating synthetic wastewater after operating for 13 months under alternating anaerobic and aerobic conditions (Yilmaz *et al.*, 2008). Moreover, the high suspended solids in the effluent of the SBR limited the overall efficiency to 68%, 86% and 74% for total COD, total nitrogen (TN) and total phosphorous (TP) respectively, in the afore-mentioned experiments (Yilmaz *et al.*, 2008). The removal of nitrogen was likely to have taken place via nitrite, thus optimizing the use of the limited COD available in the wastewater. *Accumulibacter spp.* was found to be responsible for most of the denitrification (Yilmaz *et al.*, 2008).

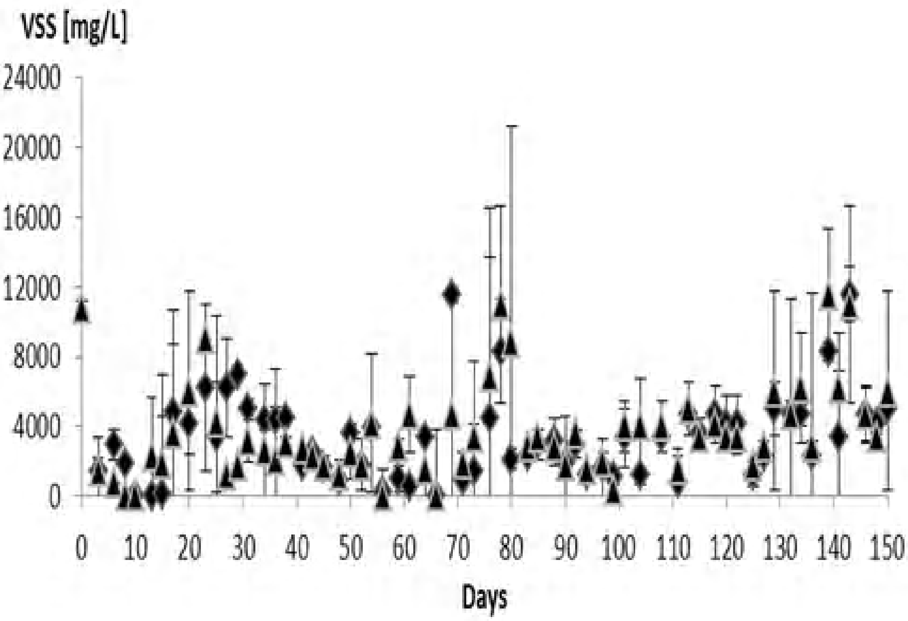

Fig. 2. Volatile Suspended Solids in the effluent of the reactors with (w-z) and without (w/o-z) zeolite. a) Recirculation (start-up or stage 1): days 0-21; b) NLR$_1$: 0.16 kg N-NO_2^-/m^3/d: days 22-53; c) NLR$_2$: 0.48 kg N-NO_2^-/m^3/d: days 54-94; d) NLR$_3$: 0.81 kg N-NO_2^-/m^3/d: days: 95-147. ♦: with zeolite; ▲: without zeolite

Table 4. Average VSS concentrations in the effluents of the modified UASB reactor

Stage	NLR (kg N-NO_2^-/m^3/d)	OLR (kg COD/m^3/d)	v_s (m/h)	VSS in effluent (g/L)
5	0.81	5.0	1.5	3.82
6	1.22	7.6	2.5	3.98
7	1.22	7.6	4.0	4.55
8	1.22	7.6	5.5	4.78

Figs 3 and 4 correspond to the balance and behavior of nitrogen species during the operation of both reactors (stages 1-4) with and without zeolite, respectively. It can be observed that the tendencies were similar in both cases, which suggests that no differences existed with respect to the utilization of zeolite in this type of reactor for the degradation of nitrite under normal operational conditions of UASB reactors. According to Fig. 3, during the recirculation period (stage 1) and stage 2 (NLR_1 = 0.16 kg N-NO_2^-/m^3/d) of the reactor with zeolite, there was neither degradation nor generation of gaseous nitrogen.The removal efficiencies of nitrogen in this reactor were 21.4% and 15.5% in stages 1 and 2, respectively. Then, during the eighth week of the study an average of 69.2% of nitrogen removal was obtained at stage 3 (NLR_2 = 0.48 kg N-NO_2^-/m^3/d), while 77.5% nitrogen removal was achieved at stage 4 (NLR_3 = 0.81 kg N-NO_2^-/m^3/d). This increase in nitrogen removal was proportional to the nitrogenous and organic loads put into the system. At the start of the tenth week, the concentration of nitrogen in the effluent began to decrease, hence the accumulation in the balance curve of Fig. 3 increasing proportionally in time and in relation to the NLR in the influent. This accumulation was due, in great part, to the gaseous nitrogen formed in the denitrification by the reduction of the nitrite present. The operational performance of the reactor without zeolite (Fig. 4) was similar to that observed in the reactor with zeolite, except during the recirculation step (stage 1) and in the step with NLR_1, where the degradation percentages of nitrite corresponded to 4.2% and 0%, respectively. Afterwards, the behavior of both reactors during the stages with NLR_2 and NLR_3 was similar. The fact that the reduction of nitrogen is greater in the reactor with zeolite demonstrated the

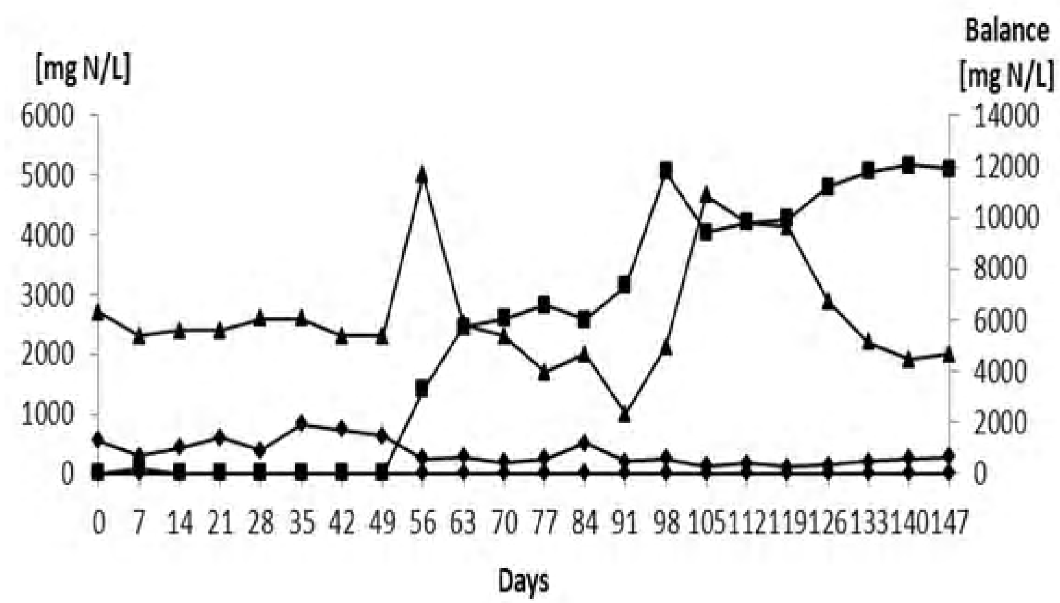

Fig. 3. Behavior of nitrogen compounds during the denitrification process via nitrite in the reactor with zeolite. a) Recirculation (start-up or stage 1): days 0-21; b) NLR_1= 0.16 kg N-NO_2^-/m^3/d: days: 22-53; c) NLR_2= 0.48 kg N-NO_2^-/m^3/d: days: 54-94; d) NLR_3= 0.81 kg N-NO_2^-/m^3/d: days: 95-147. ▲ : N-NO_2^-;• : N-NO_3^-; ◆ : N-NH_4^+; ■ %: Balance

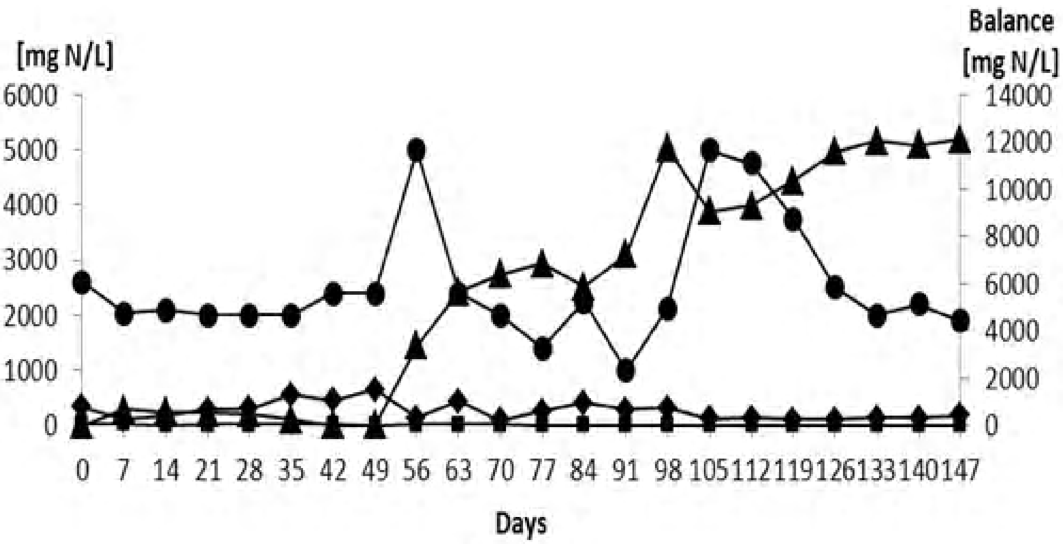

Fig. 4. Behavior of nitrogen compounds during the denitrification process via nitrite in the reactor without zeolite. a) Recirculation (start-up or stage 1): days 0-21; b) NLR_1= 0.16 kg N-NO_2^-/m^3/d: days: 22-53; c) NLR_2= 0.48 kg N-NO_2^-/m^3/d: days: 54-94; d) NLR_3= 0.81 kg N-NO_2^-/m^3/d days 95-147. • : N-NO_2^-; ◆: N-NO_3^-; ■ : N-NH_4^+ ▲: Balance

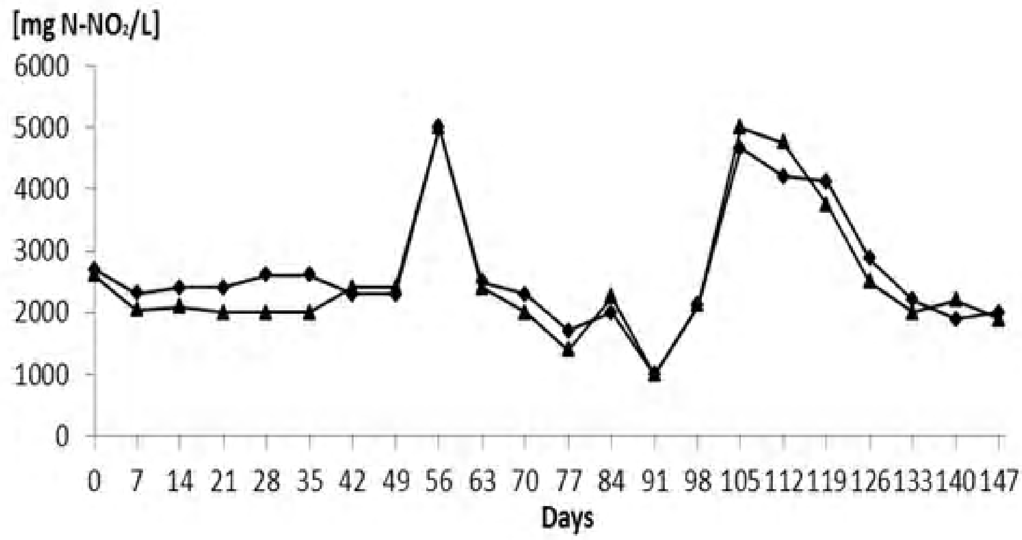

Fig. 5. Behavior of nitrite during the denitrification process via nitrite in both reactors a) Recirculation (start-up or stage 1): days: 0-21; b) NLR_1= 0.16 kg N-NO_2^-/m^3/d: days 22-53; c) NLR_2= 0.48 kg N-NO_2^-/m^3/d: days: 54-94; d) NLR_3= 0.81 kg N-NO_2^-/m^3/d: days 95-147. ▲ : with zeolite; ◆ : without zeolite

higher stability and effectiveness of the adhered microorganisms.

The concentration of nitrite (Fig. 5) decreased by 28% at stage 1 in the reactor with zeolite, while in the reactor without zeolite it only degraded by 19% (initial amount of nitrite fed was 2841 mg N/L). For an NLR_1 of 0.16 kg N-NO_2^-/m^3/d (stage 2), the degradation of nitrite increased up to 30% and 19% in the reactors with and without zeolite, respectively. For an NLR_2 of 0.48 kg N-NO_2^-/m^3/d (stage 3), the nitrite degraded in both reactors increased by up to 88% of its initial concentration (8522 mg N/L). Finally, for an NLR_3 of 0.81 kg N-NO_2^-/m^3/d (stage 4), the degradation of nitrite achieved in both reactors was 87%. Therefore, although in the first two stages (recirculation and NLR_1) the effluent nitrite concentration was lower in the reactor with zeolite, similar values were achieved in both reactors at the end of stage 2 (NLR_1). Afterwards, both reactors followed a similar pattern, without any important differences. This can be attributed, once again, to the fact that the adherence of microorganisms on the zeolite allows the biomass to obtain higher stability in less time, and therefore reducing nitrite faster during the first stages of the process at v_s less than 1 m/h, to finally achieve a similar percentage of degradation of this compound, independently of whether zeolite is used or not.

On the other hand, an NLR of 0.5 kg $N\text{-}NO_2^-/m^3/d$ - very similar to that used during stage 3 of the present work (NLR_2 of 0.48 kg $N\text{-}NO_2^-/m^3/d$) - led to an average nitrogen removal efficiency of 68% in the Anammox treatment of the effluents generated in an anaerobic digester processing wastewater from a fish cannery once previously treated in a Sharon reactor (Dapena-Mora *et al.*, 2006). These effluents contained a high content of ammonium (700-1000 g NH_4^+/m^3), organic carbon (1,000-1,300 g TOC/m^3) and salinity up to 8,000-10,000 g $NaCl/m^3$. In addition, the above-mentioned Annamox reactor showed an unexpected robustness despite the continuous variations in the influent composition regarding ammonium and nitrite concentrations (Dapena-Mora *et al.*, 2006).

Other reactor configurations such as anaerobic continuous stirred tank reactors were also used for biological denitrification at low NLR values, eliminating carbon, nitrogen and sulfur (Reyes-Avila *et al.*, 2004). In this case, NLR values of 0.2 kg $N\text{-}NO_2^-/m^3/d$ were used in the treatment of refinery wastewaters, which were characterized by their high content in ammonium, sulfide and aromatic compounds. Nitrogen removal efficiencies higher than 90% were achieved in the afore-mentioned case with a simultaneous carbon removal efficiency of 65% (Reyes-Avila *et al.*, 2004).

As was previously mentioned, only the modified UASB reactor was operated during this second experimental part due to the fact that when the v_s was higher than 1 m/h, a washing of the microorganisms was produced in the conventional UASB reactor. Fig. 6 shows the balance and behavior of nitrogenous species during the operation of the modified UASB reactor. At the beginning of the first month of experiment part II, the nitrogen concentration in the effluent began to decrease, hence the accumulation shown in the balance curve of Fig. 6 increasing proportionally in time and to the NLR of the influent. As seen in Fig. 6, stage 5 - with an NLR of 0.81 kg $N\text{-}NO_2^-/m^3/d$ - saw an increase in v_s with respect to previous stages whereas nitrite remained untouched. Then, for stages 6 to 8, where the NLR increased to

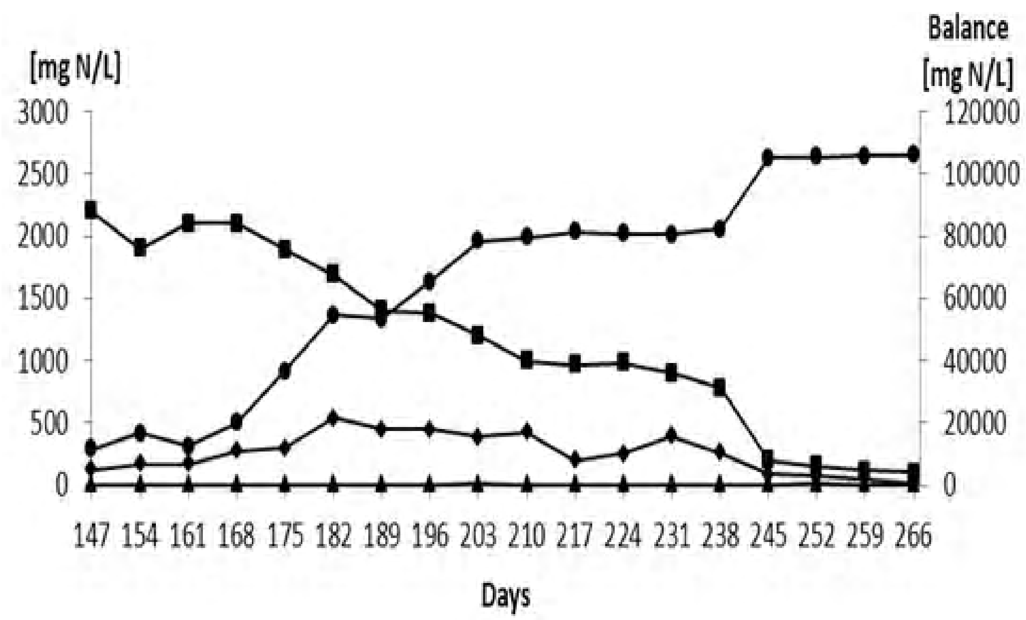

Fig. 6. Behavior of nitrogenous compounds during the process of denitrification via nitrite in the reactor with zeolite. e) NLR_4= 0.81 kg $N\text{-}NO_2^-/m^3/d$: days: 148-166; f) NLR_5= 1.22 kg $N\text{-}NO_2^-/m^3/d$: days 167-201; g) NLR_6= 1.22 kg $N\text{-}NO_2^-/m^3/d$: days 202-235; h) NLR_7= 1.22 kg $N\text{-}NO_2^-/m^3/d$: days: 236-256. ■ : $N\text{-}NO_2^-$; ◆ : $N\text{-}NO_3^-$; ▲ : $N\text{-}NH_4^+$; • : Balance

1.22 kg N-NO_2^-/m^3/d and remained constant for these three final stages with the reactor operating at a v_s of 2.5, 4.0 and 5.5 m/h, nitrogen removal efficiencies close to 50%, 65% and 95.5% were obtained, respectively. This increase was proportional to the nitrogen and organic loads entered into the system, and especially to the v_s applied to the modified UASB reactor.

Lower total nitrogen removals (48.1%-82.8%) were obtained in a combined system consisting of a UASB and an aerobic membrane bioreactor (MBR) for the treatment of low-strength synthetic wastewater (An *et al.*, 2008). The system operated at 28-30 ºC and a pH of 7.8-8.1. The lower carbon requirement for denitrification via nitrite rather than nitrate led to simultaneous methanogenesis in the same UASB reactor. A modified anaerobic baffled reactor (ABR) with eight compartments was demonstrated to be very efficient for nitrogen removal during the treatment of a synthetic sucrose/protein wastewater (Barber and Stuckey, 2000) when operated at an OLR of 4.8 kg COD/m^3/d (hydraulic retention time of 20 h), which was very similar to that used in the present work in stage 5 (5 kg COD/m^3/d). In this case, virtually all the nitrate was removed in the first two compartments, achieving denitrification efficiencies of 82% and 96% in compartments 1 and 2, respectively. Denitrification also influenced the ratio of volatile fatty acids produced and catabolized with a significant reduction in propionate and butyrate, while acetate levels increased (Barber and Stuckey, 2000). With a view to analysing the data in Fig. 6, it can be observed that during the first month the nitrite showed no important decrease when compared to previous stages (experiment part I). However, as and from stage 6, the decrease of nitrite became evident, especially at the eighth stage, where a reduction of 95.5% was obtained. Therefore, the influence of v_s on biochemical reactions is considerable, probably due to the better heat and mass transfers that occur at a v_s of 5.5 m/h. It is also interesting to note, that at the highest superficial velocity studied in this work, microorganisms were not lost (from detachment from the support and/or dragging out of the reactor), which helped achieve this high percentage of denitrification via nitrite. Slightly higher total nitrogen removal efficiencies (99%) were obtained in the treatment of real leachate from municipal landfills with high ammonium N content by using a lab-scale first-stage UASB reactor – SBR biological system (Sun *et al.*, 2010). However, in the previously mentioned case the OLR used (5.3 kg COD/m^3/d) was lower than that used in the present work operating with the modified UASB reactor during the last three stages (7.6 kg COD/m^3/d). Sun *et al.* (2009) also demonstrated that denitrification and methanogenesis were conducted in a first-stage anoxic/anaerobic UASB reactor, although the process was completed with an A/O stage. Furthermore, the NLR used in the aforementioned work (1.1 kg N-NO_2^-/m^3/d) were slightly lower than those used in the present work (1.22 kg N-NO_2^-/m^3/d) during the last three stages. Nitrate and ammonium are undesirable compounds, but were nevertheless found in the effluent of the modified UASB reactor at very low concentrations during the last stages of operation. Moreover, these concentrations were much lower than those found in final effluents of other denitrification processes (Dapena-Mora *et al.*, 2006; An *et al.*, 2008).

CONCLUSION

The modified UASB reactor was very effective operating at superficial velocities (v_s) of up to 5.5 m/h, a value much higher than the superficial velocity achieved in the conventional UASB (1 m/h). At a v_s over 5.5 m/h, the hydraulic behavior of the reactor was similar to that of an expanded bed since the sludge permanently occupied between 40% and 50% of the reactor's volume. When the superficial velocities were less than or equal to 1 m/h, there were no significant differences in the general behavior of the two reactors. The only noteworthy observation was the higher initial velocity of denitrification achieved when zeolite was used. However, an almost identical final denitrification percentage (87%) was achieved in each one of the first stages studied (stages 2 to 4), which differed slightly in v_s and NLR.

The concentration of VSS in both reactor effluents was similar during stages 1 to 4 (lower than 3.7 g/L), which leads to the conclusion that when the v_s is found in the typical ranges of a UASB (less than 1 m/h), there was no difference with respect to whether the microorganisms were found in granule form or adhered to a support. In the modified UASB, at v_s higher than 1 m/h, no detachment and/or dragging of microorganisms from the reactor were observed because the concentrations of VSS in the effluents were never higher than 4.8 g/L. This concludes the suitability of operating at these v_s ranges (up to 5.5 m/h). Nitrite removal efficiency during the first part of the experiment (v_s less than 1 m/h) was slightly higher in the modified UASB reactor with zeolite, which favored microorganism activity and process performance, shortening the start-up period and the time necessary to achieve adequate stability.

The removal of nitrite increased significantly when v_s was higher than 1 m/h, to such an extent that operating at a constant NLR of 1.22 kg N-NO_2^-/m^3/d, a nitrogen removal efficiency close to 50% was obtained for a v_s of 2.5 m/h, 65% at a v_s of 4.0 m/h and 95.5% at a v_s de 5.5 m/h. This last value demonstrated the great advantage of the use of the modified UASB reactor operating at v_s as high as 5.5 m/h.

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