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Comparison of different strategies for nitrogen removal by simultaneous nitrification and denitrification process in a batch rotating disk reactor

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ABSTRACT: *In this paper, different strategies of aeration and organics feed in the reactor were studied to evaluate the simultaneous nitrification and denitrification (SND) process efficiency for a sequencing batch rotating disk biofilm reactor (SBRDR). The effect of C/N ratio on biological nitrogen removal was also studied. Among the four strategies used, the ones with organic feeding showed the best results to promote SND. It was also observed a different behavior: the efficiency of SND as a function of the C/N ratio shows a tendency to a have a lower value than values previously reported in the literature. A continuous or intermittent aeration in the process did not improved the nitrogen removal. These results provide sufficient knowledge of the parameters that have the most influence on the efficient nitrogen removal via SND in batch rotating disk reactors.*

Keywords: *batch reactor,denitrification, nitrification, nitrogen removal, rotating disk.*

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I. INTRODUCTION

The biological elimination of nitrogen in wastewater treatment plants results from the processes of nitrification and denitrification. These two processes have been thought to be two subsequent reactions by different groups of microorganisms. Aerobic autotrophic ammonia oxidizer bacteria (AOB) oxidize ammonia to nitrite, afterwards nitrite oxidizer bacteria (NOB) oxidize nitrite to nitrate. Under anoxic conditions, nitrite and then nitrate are reduced to molecular nitrogen by heterotrophic denitrifying bacteria [1].

Simultaneous nitrification and denitrification (SND) happens when these two biological reactions occur at the same time in the same reactor [2]. This process is based on the oxidation of ammonium mainly up to nitrite by limiting the activity of NOB [3]. The main physical explanation is that SND occurs within microbial flakes as a result of low dissolved oxygen (DO) concentrations arising from diffusional limitations. Therefore, there exists anoxic microzones in the center of the sludge flakes or in the inner parts of biofilms that allow heterotrophic to denitrify NO_X^- to molecular nitrogen [4].

When SND goes along with the inhibition/limitation of the NOB growth (nitrification step), a novel process of nitrification-denitrification via nitrite leads to many advantages over conventional SND such as: lower oxygen consumption during nitrification (25%), less need of organics for denitrification (40–60%), lower sludge production, shortened reaction time (25%) and no apparent nitrite toxicity effects for the microorganisms in the reactor [2]. However, the difficulty in removing nitrogen via nitrite lies in achieving specific ammonia inhibition and oxygen limitation in order to washout the NOB from the reactor without strong retreating in the ammonia oxidation rate [2].

From previous studies, it was found that there are a lot of factors which influence SND, such as structure, size, density and concentration of sludge flakes [5, 6], DO [4, 6, 7], food/microorganism (F/M) ratio [1], carbon/nitrogen (C/N) ratio [7-9] and pH [10, 11]. However, there are several factors that have not yet been explored, such as strategies of aeration and feed of organics on the SND efficiency.

Pochana and Keller [5] have investigated the efficiency of nitrogen removal from wastewater by an SND-based sequencing batch reactor (SBR). They have observed that higher DO concentrations enhance the nitrification rates. Simultaneously, high DO concentrations inhibit the denitrification process, causing an accumulation of nitrite and nitrate in the reactor. On the other hand, at lower DO concentrations the nitrification processes is inhibited and the denitrification process is enhanced. Therefore, the DO level is a critical factor to the SND process.

The C/N ratio is also an important factor, since competition of substrates, i.e., organic carbon and ammonia, for oxygen among bacterial species occurs within a biofilm; the basic problem in nitrification lies in the fact that nitrifying bacteria have much lower growth rate and higher affinity to oxygen than those of heterotrophic bacteria. Thus, nitrifying bacteria can be surmounted by heterotrophic bacteria for dissolved oxygen (DO), what leads to the activity of the nitrifying bacteria the innermost area of the biofilms, where DO concentration is lower than in the outermost area [8]. Since autotrophic AOB have generally been characterized by low growth rates and poor yields, the nitrification is generally a rate-limiting step in a biological nitrogen removal process [1].

Meng et al. (2008) [7] have studied how the C/N ratio controls the simultaneous nitrification and denitrification process using an airlift internal circulation membrane bioreactor. They have proposed that the system can achieve nearly complete removal of both organic matter and NH_4^+-N, with no accumulation of NO_2^--N and NO_3^--N, when the C/N ratio is controlled at 10.04 and 15.11. Moreover, an increase in the rate COD/N can also harm the nitrification step, decreasing the efficiency of removal of nitrogen, as observed in the work of Zou et al. (2012) [9]. Matsumoto and Tsuneda (2007) [8] reports that the efficient simultaneous C and N removal -more than 80% and 70%, respectively- was achieved at the range of C/N ratio from 3.0 to 5.25. Maximum total nitrogen (T-N) removal efficiency is as high as 78.9% at a C/N ratio of 3.75.

The feed procedure in the reactors most frequently found in the literature include the addition of organic matter in the aerobic and anoxic step [4, 7, 12]. For the aeration, strategies with fixed aeration [7, 13] and intermittent ones [2, 12] are found in the literature.

In this sense, due to the range of values found in the literature, the aims of this work were:

-to evaluate different strategies of aeration and feed of organics on the SND efficiency for a sequencing batch rotating disk biofilm reactor (SBRDR)

-to evaluate the effect of C/N ratio on biological nitrogen removal.

II. MATERIALS AND METHODS

The experimental apparatus consisted of an acrylic reactor with a rotating disk (35.4 cm. diameter) inoculated with nitrifying and denitrifying bacteria, i. e. AOB, NOB and heterotrophic bacteria groups. The disk is kept completely immersed in a volume of 6 liters of effluent controlled by a programmable logic controller operated by Matlab 7.1. The monitoring and control system consisted of oxygen and pH/temperature electrodes (WTW Oxi 701, Germany, and HACH EC 310, USA, respectively). The electrodes were calibrated every 2 weeks and before set-point change. The 0.5M sodium carbonate solution used for H+ neutralization during nitrification was dosed by a diaphragm pump (LANG, type ELADOS EMP II, 41 L/h, Germany), and the aeration was carried out by using pulse width modulation for electromagnetic valve opening (Festo 457, MSG-24DC, Germany). The air was supplied by an aquarium aeration pump (COSMOS double type 1000, China). The data for pH, temperature, DO concentration, and the pumped carbonate solution volume were acquired, processed, and stored every 5 min by means of a Programmable Logic Control (PLC) (Siemens, Simatic S7-200) connected to an IBM-compatible PC, as can be seen in Fig. 1. The pH/temperature and DO signals were transmitted to the PLC by the local devices described above. Through specific sensors it was generated a record of real-time monitoring of pH, DO, temperature and consumption of carbonate. With these information, the program automatically determined the duration of each cycle. Operating parameters such as periods of aeration, feeding, the addition of carbon sources were automated and also controlled by this software.

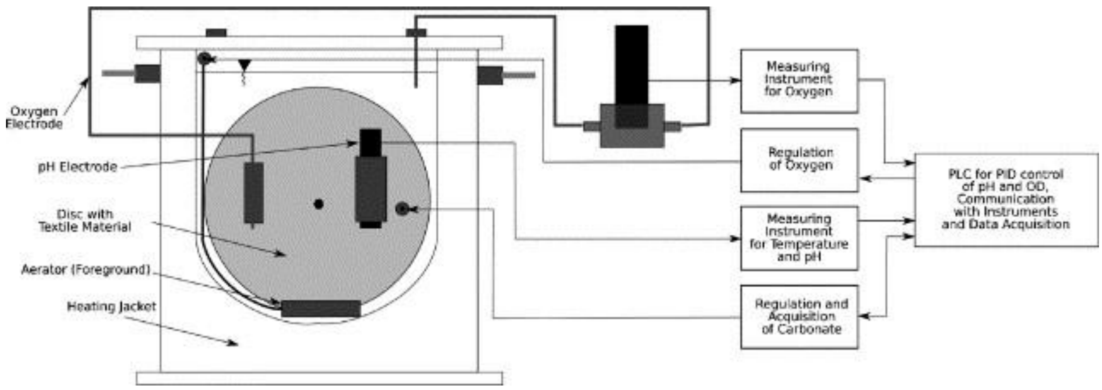

Figure 1 Instrumentation and control system [3].

For all operations, the reactor was fed with a synthetic medium with approximately 150 mgNH_4-N/L as initial total ammonium nitrogen (TAN) concentration, according to previous studies[3]. All substrates were prepared with a constant ratio of N:P:Mg = 340:10:1. The four operating strategies are described in Table 1. All strategies operate on SBR mode. The aeration was suspended in anoxic phase and there was addition of carbon source, that means an acetate solution with a chemical oxygen demand (COD) of 10,000 mgO_2/L.

Table 1. Operating strategies used on Rotating Disk Reactor.

Strategy	Feed in AerobicStage	AerationForm	DO in Aerobic Stage (mg/L)	COD/N	Operation Time (days)
Run I	Without COD	Continuous	1.5 and 2.0	-	217
Run II	With COD	Intermittent	1.0/2.0	2, 3 and 4	241
Run III	Pulses of COD	Intermittent	1.0/2.5	-	209
Run IV	With COD	Continuous	2.0 and 5.0	2	68

*COD means the acetate solution (carbon source).

On Run I carbon source was added only in denitrification. Run II e IV had feedings with COD (acetate solution) and Run III had COD addition when the DO concentration was at the minimum aeration set point. The use of intermittent aeration with short cycles can allow the nitrification and denitrification occurrence at the same time, especially at the start of the aeration period, when the low DO concentration is not inhibiting the nitrification [12, 14-16].

Intermittent aeration was used in the aerobic phase, which consisted of periods of higher DO concentration and periods with lower DO concentration. When the DO concentration reached the minimum value set for each strategy, the system added pulses of organic matter, as a mean to promote denitrification of the nitrite molecules generated by AOB during the higher DO concentration. Furthermore, the increase of COD in the period of low DO concentration prevents that organic matter is oxidized at the points of highest DO concentration. Using the information pH in the homogeneous liquid, the system determined the end of the aerobic phase. When starting the anoxic phase, the aeration was stopped and COD pulses were added to the reactor. Also by pH measurement, the end of the cycle was detected. At the end of each cycle, the reactor was manually emptied and refilled with synthetic medium with 150mgNH_4-N/L .

TAN concentration was measured using an ion-selective electrode (Thermo Orion 95-12, 2001, USA); nitrite and nitrate were measured using standard spectrophotometric methods [17]. It was evaluated if there is a statistical difference between the strategies with respect to the percentage of nitrogen removal by SND, with a confidence interval of 95%, using the software Statgraphics Centurion XVI.

III. RESULTSANDDISCUSSION

In Fig. 2 it is possible to evaluate the profiles obtained for each strategy in this work.

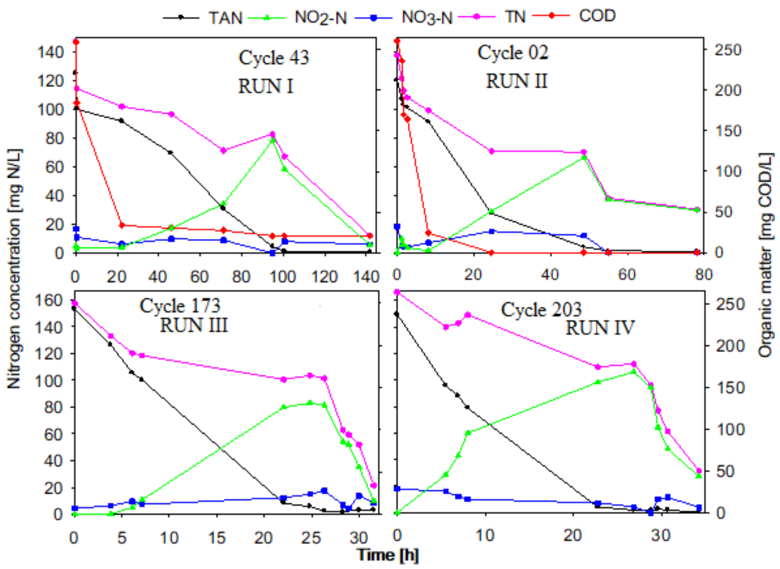

Figure 2. Concentration profile of the nitrogen species and carbon source of the four operating strategies.

As it is possible to observe, the four “runs” promoted SND. There was accumulation of nitrite in the aerobic step and this was consumed in the anoxic stage. The nitrate concentrations obtained were lower than 20 mg/L. The four operate strategies promote SND, however, with the repetition of cycles, there was significant differences between them, which will be presented below.

3.1 Statistical analysis about nitrogen removal by SND

A statistical analysis between the strategies, with respect to the percentage of nitrogen removal by SND, was processed with the program Statgraphics Centurion XVI, with a confidence interval of 95%. It was found that there is a statistical difference between: Run I and Run II; Run I and Run III; Run I and Run IV; Run II and Run IV; Run III and Run IV. There is no significant difference between Run II e Run IV. Fig. 3 presents the results for SND and nitrite accumulation in the four strategies.

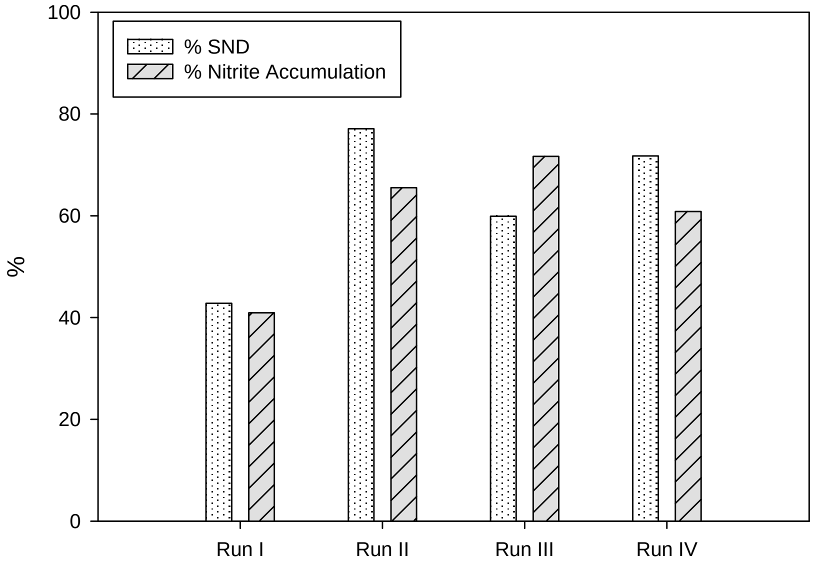

Figure 3.SND efficiency and percentage of nitrite accumulation in different strategies of operation, in a Batch Rotating Disk Reactor.

` Among the four strategies, the ones that result in a higher nitrogen removal by SND were Run II and IV, followed by Run III and Run I. Regarding the form of aeration, Run II was operated with intermittent aeration and Run IV with continuous aeration. Despite this difference in the process, the two treatments showed no significant differences on the efficiency of nitrogen removal by SND. What is similar between them is the feeding strategy (with COD). Apparently, the addition of COD in the aerobic phase, regardless of the aeration strategy, is more important to the oxidation of NOx compounds produced in the aerobic phase. Intermediate values between the best result (Run II and IV) and worst (Run I) were obtained in Run III, where the feeding strategy is the COD Pulse, also a form of intermediate feed between "with COD" and "without COD".

For the accumulation of nitrite, significant differences were observed between Run I and Run II, as well as between Run I and Run III. The differences between Run I and Run II are the aeration and the feed procedures .

Using a polyurethane packed-bed-biofilm batch reactor, Daniel et al. (2009) [18] studied the removal of ammonium via SND with intermittent aeration strategy. DO concentrations were kept within the range of 2.0– 2.7 mg/l and ethanol was used as an electron donor for the denitrification phase at final carbon to nitrogen ratios of 3:1. They observed that a consistent decrease of nitrite concentration started always immediately after the interruption of oxygen supply and addition of the electron donor. The results showed that nitrogen removal efficiencies of dissolved forms were higher than 99% throughout the trials. It was observed a synchronized variation between nitrite and dissolved oxygen concentrations without a significant accumulation of nitrate. In this study there was also no significant nitrate accumulation. These results were considered a strong evidence of a dynamic and robust balance between the nitrification and denitrification nitrite-shortcut process.

In Table 2 it is possible to observe the results obtained in this work compared to the results obtained by various authors for nitrogen removal by SND using different operating parameters.

Table 2. Results found in literature compared to the ones obtained at this work about nitrogen removal by SND.

	SND (%)	COD/N	NH_4^+ oxidation (%)	NO_xaccumulation (%)	DO concentration (mg O_2/L)	Aerationstrategy
Run I	42.8	NR	96.7	40.9	1.5 and 2.0	continuous
Run II	77.1	2, 3 and4	62.3	65.5	1.0/2.0	intermittent
Run III	59.9	NR	83.5	71.7	1.0/2.5	intermittent
Run IV	71.7	NR	76.3	60.8	2.0 and 5.0	continuous
Ciudad et al. 2007 [19]	NR	NR	NR	80	0.6-5.0	
Holman et al. 2005 [4]	75	20	NR	NR	0.0-1.6	continuous
Do Canto et al. 2008[20]	NR	2	97	NR	2.0	intermittent
Meng et al. 2008[7]	maximum	NR	NR	NR	1.0	continuous
Guoet al. 2009 [6]	44.9	3,6	NR	> 95	0.4-0.8	continuous
Daniel et al. 2009 [18]	maximum	3	NR	NR	2.0-2.7	intermittent
Yang et al. 2011 [2]	NR	5	NR	79.4	NR	intermittent

* DO values correspond to the aerobic phase. In all cases, the aeration was suspended in anoxic stage. NR corresponds to data not reported.

It is possible to notice that there is variability in the results reported in the literature. Daniel et al, (2009) [18] achieved maximum efficiency in the process of SND with intermittent aeration, higher values than the Run II and III (77.1% and 59.9% respectively, using a packed-bed batch reactor. Holman et al (2005) [4], reached 75% of SND with continuous aeration (DO of 0.0-1.6 mg / L), similar to that of Run IV which reached 71.7% (OD 2.0 and 5.0 mg/L) and worse result than Meng et al. (2008) [7], who obtained maximum SND with continuous aeration with DO equal to 1.0 mg/L using a membrane bioreactor.

Even with different aeration strategies, the oxidation of run I ammonia was comparable to the system used by Do Canto et al. (2008)[20]. The advantage of Run I would be the energy savings associated with the denitrification process, since 42.8% was via SND, whereas Do Canto et al. prioritized the conventional process.

3.2 Influence of DO concentration

Figure 3 shows that a higher DO concentration in the aerobic phase increases the ammonium oxidation rate (ex: Run I). The ammonium oxidation rate is expressed in terms of kg of total ammoniacal nitrogen oxidized per cubic meter per day.

On run IV it was possible to evaluate de DO effect on ammonium oxidation rate but no statistical differences was observed on SND efficiency. It can be seen that the DO concentration in the aerobic phase had a positive influence on the ammonium oxidation rate, but had no significant effect on the percentage of nitrogen removal by SND.

In the work of do Canto (2008) [20], also using a sequencing batch biofilm reactor, the best cycle occurred with continuous feeding. The two aeration strategies were employed (continuous and intermittent) and the best results were obtained with intermittent aeration with DO concentration equal to 2.0 mg/L.

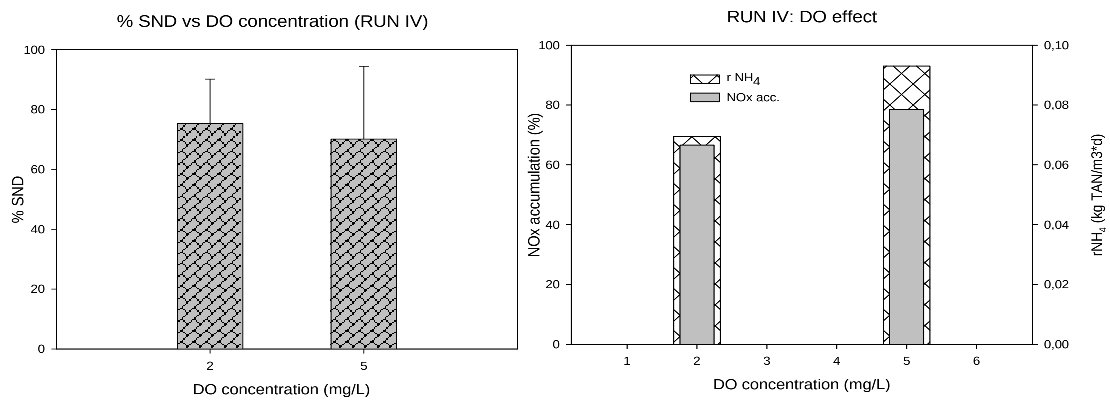

Figure 3. DO effect in Run IV for %SND, NOx accumulation and ammonia oxidation rate. “NOx acc.” means NOx accumulation.

About the nitrite accumulation on different DO concentrations, there are no statistically significant differences between any pair of trials at the 95.0% confidence level. As it is known, there are limitations in the transport of oxygen through the biofilm and probably because of this, it can not be perceived a difference in the overall process. Nevertheless it is possible to observe an improvement in the oxidation of ammonium.

SND has also been studied in bioelectrochemical systems by Virdis et al (2011) [21]. The results presented on their work show a maximal nitrogen removal efficiency of 87% at a dissolved oxygen (DO) level of 5.73 mg/L. The DO levels used in their study are higher than the thresholds previously reported as detrimental for denitrification [21]. This is an example of a process that provides SND with high DO concentrations.

3.3 Influence of COD/N in nitrogen removal by SND

Figure 4 presents the results of Run II, in which different COD/N ratios were evaluated in order to analyze which one promoted the better SND. In general, increasing the amount of organic matter in the feed improves the growth of heterotrophic bacteria in the biofilm surface competing for space with autotrophic bacteria. This will provides a decrease in the efficiency of ammonium oxidation.

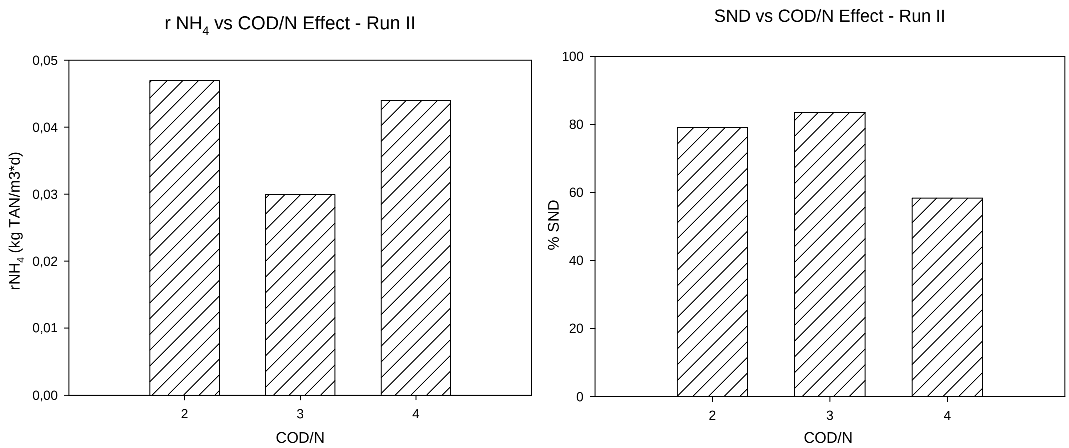

Figure 4.COD/N effect of SND and ammonium oxidation rate. The data shows only results of the Run II.

It can be observed that a higher proportion of COD/N decreased the percentage of nitrogen removal by SND. But by the data presented, this behavior is not clear, since the ratio COD/N 2 and 3 have approximately the same values. It was also observed a decrease in the ammonium oxidation rate when COD/N is 3. However the obtained values were virtually the same when COD/N is 2 or 4.

Studies have shown that in SND process in rotating disk reactors, with relations COD/N greater than 1.5, the heterotrophic biomass dominates the outer layers of the biofilm, while autotrophic biomass dominates the inner layers [14]. Cheng and Chen (1994) [22] investigated the effects of varying the COD/N-NH_4^+ ratio in a nitrifying fluidized bed reactor. The COD/N-NH_4^+ ratio was varied from 0 to 10. The increment of the C/N ratio resulted in an increase of nitrite concentration and a reducing amount of oxidized ammonia, showing a competition between the bacteria that oxidize nitrite and carbon. According to Aesoy et al (1998) [23], the COD/N ratio required for nitrogen removal using ethanol as carbon source is 4.5. Performing denitrification via nitrite, Abeling and Seyfried (1992) [24] and Katsogiannis et al. (2003) [25] worked on relations COD/N of 2.8 and 3 respectively.

In the work of Meng et al. (2008) [7], in the COD/N ratio of 4.77 and 10.04, the system nitrogen removal efficiency became higher than 70%. However, the nitrogen removal efficiency decreased to less than 50%, as the COD/N ratio shifted to 15.11.

As it can be seen, there is much variation in the COD/N ratios turning the choice of an optimal proportion a difficult step. In the case of Run II, the proportion 2 and 3 are more appropriate for promoting SND. The values obtained reached approximately 80% of SND.With a ratio of 2 and 4 it was possible to promote a comparable ammonium oxidation rate. Therefore, in Run II, the most appropriate rate of COD/N was 2.

IV. CONCLUSION

The four operate strategies promote SND and there was significant differences between them. The best strategies to promote SND were Run II and Run IV.On run IV it was possible to evaluate de DO effect on ammonium oxidation rate but no statistical differences was observed on SND efficiency. It can be seen that the

DO concentration in the aerobic phase had a positive influence on the ammonium oxidation rate, but had no significant effect on the percentage of nitrogen removal by SND.

A higher proportion of COD/N decreased the percentage of nitrogen removal by SND. The behavior observed in the efficiency of SND as a function of the COD/N ratio shows a tendency to a lower value than values previously reported in the literature.

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