DEAMMONIFICATION ACTIVITY IN DIFFERENT REACTORS CONFIGURATIONS SUBMITTED TO HIGH RECIRCULATION RATES

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SUMMARY: Deammonification process is being studied as an alternative for digestate post treatment. In this process, the ammonia oxidation by partial nitrification generates substrate to anammox activity, so there must have partial nitrification control to prevent nitrite accumulation. Nitrifying bacteria (aerobic microorganisms) and anammox (anoxic microorganisms), have to cooperate to complete ammonia removal, what makes nitrifying bacteria growth an important key to process control. Therefore it is necessary a strict control of nitrite production that can inhibit the anammox process. Nitrite concentration in the reactor can be controlled at different strategies, such as aeration, reactor configuration and recirculation rate. Considering this, the present study aimed to evaluate the effect of high recirculation rates during deammonification process in two reactor configurations: EGSB and Imhoff cone. The high recirculation rates improve the mass transfer in the reactors, but also accelerate nitrifying growth, making clear that it is not a good strategy to be used in deammonification process.

Keywords: Anammox, nitrogen removal, partial nitrification.

INTRODUCTION

Recent studies have been developed aiming to improve treatment efficiency and to reduce operational costs. It is possible by optimizing the treatment strategies available or pursuing development of new processes capable to remove high wastewater nitrogen loads (Casagrande, et al. 2013). Several alternative processes were developed in recent years, such as partial nitrification (PN), anaerobic ammonium oxidation (anammox) and their combined system (deammonification) (Vanotti, et al. 2013).

Deammonification is a completely autotrophic nitrogen removal process in which nitrifying bacteria (aerobic microorganisms) and anammox (anaerobic microorganisms) have to cooperate (Magrì, et al. 2012). Thus, the ammonia oxidation by PN generates substrate to anammox activity. The process can be chemically represented by the Equation 1 (Sliekers, et al. 2002; Vanotti, et al. 2012).

\[
\text{NH}_3 + 0.85\text{O}_2 \rightarrow 0.11\text{NO}_3^- + 0.44\text{N}_2 + 1.14\text{H}^+ + 1.43\text{H}_2\text{O} \quad \text{Equation 1}
\]

For deammonification success PN must be controlled, preventing nitrite accumulation and the possible nitrite oxidation to nitrate by nitrite-oxidizing bacteria. Thus, nitrifying bacteria growth is an important key to control and stabilization of the process (Chung, et al. 2005). Anammox bacteria have a cell duplication rate around 0.09 d⁻¹ and ammonium oxidizing bacteria (AOB) 0.801 d⁻¹ (Hellinga, et al. 1998; Strous, et al. 1998).

One of the strategies to control bacteria growth is using different reactor configurations. In this way, few studies reported that up-flow reactors can eliminate dead zones and improve mass transfer, by expanding the granular sludge bed which can be achieved through high recirculation rates (Zheng, 2013).

In this way, high recirculation rates implies in higher superficial liquid velocity that can cause sludge bed expansion and turbulence. So, this sludge bed expansion proportionate a better mix between nitrifier and anammox bacteria, better contact with substrate and avoid dead zones (kato, 1994).
The idea is to permit a good biomass contact with the substrate and oxygen, during oxic phase, and create good anoxic conditions for anammox activity (Fang, et al. 2011).

Considering this, the present study aimed to evaluate the effect of high recirculation rates during deammonification process in two reactor configurations: EGSB and Imhoff cone.

**MATERIAL AND METHODS**

An Imhoff cone reactor (1L) containing: nitrifying bacteria biomass (r-NH₃-N= 69.8 mgN.L⁻¹.h⁻¹) and anammox bacteria biomass (r-NH₃-N= 18.4 mgN.L⁻¹.h⁻¹; r-NO₂⁻-N= 28.9 mgN.L⁻¹.h⁻¹).

An EGSB reactor (1L) containing: nitrifying biomass (r-NH₃-N= 100.4 mgN.L⁻¹.h⁻¹) and anammox biomass (r-NH₃-N= 19.5 mgN.L⁻¹.h⁻¹; r-NO₂⁻-N= 28.7 mgN.L⁻¹.h⁻¹).

These two reactors containing biofilm plastic carrier (55g w⁻¹), were operated at HRT of 6.3 h and recirculation flow rate of 43 L.d⁻¹, intermittent aeration in cycle of 30 minutes (22 min on / 8 min off) at 25°C, fed during aeration time with synthetic medium containing 300 mgNH₃-N.L⁻¹ and nutrient solution (Magri, et al. 2012). The reactors were operated during 16 days, with nitrogen loading rate (NLR) of 0.80 ± 0.02 kgN.L⁻¹.d⁻¹.

Imhoff cone reactor was inoculated with 9.50E+07 gc of nitrifying bacteria and 4.73E+09 gc of anammox. EGSB reactor was inoculated with 1.05E+08 gc of nitrifying bacteria and 4.65E+09 gc of anammox.

The N-compounds and alkalinity were daily measured according to Apha (2012). pH (Hanna, pH 21) and DO (YSI 55) were monitored at aerobic and anoxic phases.

Quantification of nitrifying and anammox bacteria were performed by qPCR, using primers described by Braker et al., (1998) and Schmid et al., (2008), respectively. qPCR reactions were performed using ΔΔCq comparation.

**RESULTS AND DISCUSSION**

In the imhoff cone reactor, nitrogen removal rate (NRR) was 0.20 ±0.12 kgN.L⁻¹.d⁻¹ and Total Nitrogen (TN) removal medium efficiency of 23.5 ±14.5%. After day 5 (Fig. 1A) nitrite started to accumulate in the reactor, indicating the influence of nitrification process.

In addition (Tab.1), after 5 days of operation, nitrifying bacteria community increased 1 log₁₀ and at 16th day increased 2 log₁₀. Anammox bacteria decreased 1 log₁₀ and at 14th day decreased more 1 log₁₀.

On the other hand, in EGSB reactor NRR was 0.35 ±0.15 kgN.L⁻¹.d⁻¹ achieving NT removal efficiency of 42 ±16%. Until 6th day, deammonification process was dominant in the reactor. That can be confirmed by low NH₃-N, NO₂⁻-N and NO₃⁻-N in the effluent (Fig. 1B). So, the reactor proved to be a good configuration to this process.

However, as in the Imhoff cone reactor, as from day 7th, nitrite started to accumulate in the reactor indicating the influence of nitrification process.

After the 5th day nitrifying bacteria community decreased 2 log₁₀ and anammox bacteria community increased 1 log₁₀. After the 16th day, nitrifying bacteria community increased 4 log₁₀, but anammox bacteria decreased 3 log₁₀ (Tab.1).

The nitrifying growth can be seen at Figure 2 at 1th, 5th and 16th day. Furthermore, EGSB reactor had greater ammonia removal and nitrite production than Imhoff cone reactor, proving that high recirculation rates provide better mass transfer, but have an influence at nitrifying growth.

Thus, proving nitrifying growth at reactors, this possible happens due to high recirculation rate that contribute to a better oxygen mass transference to nitrifiers bacteria increasing its activity and growing.
Figure 3 shows the flocs characteristics at 1st and 16th day. In the beginning of experiments flocs are structured and granular, however at the end of the experiment they are fluffy, a characteristic form of nitrifier bacteria. This can be another result of high recirculation rates that destructured this granules, thereby decreasing anammox activity which favored nitrifiers growth.

EGSB configuration have high potential to removal high nitrogen loads (Fang, et al. 2011), and also proved to be a good reactor configuration to use deammonification process. But it is necessary to change the original configuration, once that the high recirculation flow rate could accelerate nitrifying bacteria growth.

CONCLUSIONS

EGSB reactor are more resistant to nitrifiers excessive growing compared to Imhoff cone reactor when submitted to high recirculation rate. Due to an excessive oxygen transfer to the reactors, high recirculation rates are not a good strategy to be used in deammonification process.

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REFERENCES

Figure 1. Influent and effluent nitrogen concentration during operation time. A. Imhoff cone reactor B. EGSB reactor.

Figure 2. Nitrifying growth in the reactors at 1st, 5th and 16th day. A. Imhoff cone reactor B. EGSB reactor.

Figure 3. Flocs details in each reactor at 1st day and 16th day. A. Imhoff cone reactor B. EGSB reactor.

Table 1. Nitrifying and anammox bacteria quantification of Imhoff cone and EGSB reactors by qPCR

<table>
<thead>
<tr>
<th>Day</th>
<th>Imhoff cone reactor</th>
<th>EGSB reactor</th>
</tr>
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<tbody>
<tr>
<td></td>
<td>Nitrifying</td>
<td>Anammox</td>
</tr>
<tr>
<td>Inoculum</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>9.50E+07</td>
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