Water Research 104 (2016) 292–302

Contents lists available at ScienceDirect

Water Research

journal homepage: www.elsevier.com/locate/watres

The inhibitory effects of reject water on nitrifying populations grown at different biofilm thickness

Maria Piculell [a, *], Carolina Suarez [b], Chunyan Li [c], Magnus Christensson [a], Frank Persson [d], Michael Wagner [c], Malte Hermansson [b], Karin Jönsson [e], Thomas Welander [a]

[a] Veolia Water Technologies AB – AnoxKaldnes, Klosterängsvägen 11A, SE-226 47 Lund, Sweden
[b] Dept. of Chemistry and Molecular Biology/Microbiology, University of Gothenburg, Medicinaregatan 9E, SE-413 90 Gothenburg, Sweden
[c] Water Chemistry and Water Technology, Engler-Bunte-Institut, Karlsruhe Institute of Technology, Engler-Bunte-Ring 9, 76131 Karlsruhe, Germany
[d] Water Environment Technology, Dept. of Civil and Environmental Engineering, Chalmers University of Technology, SE-412 96 Gothenburg, Sweden
[e] Water and Environmental Engineering, Dept. of Chemical Engineering, Lund University, P.O. Box 124, SE-221 00 Lund, Sweden

ARTICLE INFO

Article history:
Received 27 April 2016
Received in revised form
12 August 2016
Accepted 13 August 2016
Available online 15 August 2016

Keywords:
Biofilm thickness
FISH-CLSM
MBBR
NOB inhibition
Nitritation-anammox
Optical coherence tomography

ABSTRACT

Suppression of nitrite oxidizing bacteria (NOB) is of vital importance to achieve successful, energy efficient, mainstream anammox processes for wastewater treatment. In this study, biofilm carriers from a fully nitrifying MBBR system, fed with mainstream wastewater, were temporarily exposed to reject water from sludge dewatering, to evaluate this as a possible strategy to inhibit NOB and achieve nitrite production under realistic conditions. Two different carrier types were compared, in which biofilm thickness was maintained at approximately 400 and 50 μm, respectively, and reject treatment was tested at different exposure time and loading rates. Reject exposure almost always resulted in an increased nitrite production in the thinner biofilm, and overall, nitrifiers growing in the thin biofilm were more sensitive than those grown in the thicker biofilm. The effect from reject exposure remained in the systems for four days after returning to mainstream operation, with nitrite production gradually increasing for three days. Increased concentrations of free ammonia correlated with reject exposure and may be the cause of inhibition, although other factors cannot be excluded.

1. Introduction

Nitritation refers to the first step of nitrification, i.e. the oxidation of ammonium to nitrite by autotrophic ammonia-oxidizing bacteria (AOB), and is the key for achieving alternative, energy efficient nitrogen removal processes such as partial nitrification and anammox (PNA). In conventional nitrogen removal, AOB and nitrite-oxidizing bacteria (NOB) co-exist, and ammonium is completely oxidized to nitrate, which is then reduced to nitrogen gas in the denitrification process. In PNA systems, however, the oxidation of ammonium is instead halted at nitrite which is converted to nitrogen gas by anammox bacteria. Hence, PNA requires less oxygen and carbon compared to conventional processes, and reduces the overall energy requirements for nitrogen removal (Daigger, 2014). The activity of NOB is detrimental in PNA systems, as NOB will compete with AOB for oxygen and with anammox for nitrite. A successful suppression of NOB is therefore of vital importance when applying PNA processes in wastewater treatment (Al-Omari et al., 2015; Xu et al., 2015).

The suppression of NOB relies on a competitive advantage of AOB over NOB, which in turn depends on the oxygen affinity, growth rate, temperature and resilience of the two bacterial groups towards inhibiting compounds, such as high levels of free ammonia (FA) or free nitrous acid (FNA). There are several examples of successful nitritation, especially for PNA processes applied in side-stream treatment of digester centrate ("reject water") (Lackner et al., 2014), operating at high temperatures (>20 °C), low oxygen concentrations (<2 mg/L) and high ammonium concentrations (>100 mg/L). However, NOB suppression becomes considerably more challenging when applying PNA processes in more diluted wastewaters at lower temperature, such as municipal mainstream water. There are operational approaches to suppress NOB at mainstream conditions, for example by operating at low dissolved oxygen (DO) and high effluent ammonium concentrations, to ensure higher growth rate of AOB over NOB (Isanta et al., 2015).

* Corresponding author.
E-mail address: maria.piculell@anoxkaldnes.com (M. Piculell).

http://dx.doi.org/10.1016/j.watres.2016.08.027
0043-1354/

However, both AOB and NOB activities are strictly oxygen dependent (Rusten et al., 2006), and oxygen limitation will also limit the nitritation rate, hence affecting the overall efficiency of the PNA process.

An alternative approach to suppress NOB in mainstream operation is to regularly alternate between mainstream and reject operation, either by moving the biomass or by switching the feed, in order to expose biomass to favorable conditions for AOB growth (Lemaire et al., 2014; Wang et al., 2014). A recent finding by Piculell et al. (2016b) indicated that NOB suppression could be achieved at high DO in a nitrifying Moving Bed Biofilm Reactor (MBBR), provided that the biofilm thickness was limited, below 300 μm. It was hence suggested that PNA could be achieved in a two-stage configuration, with nitritation and anammox growing in consecutive MBBRs. By combining an alternating feeding scheme with a limited biofilm thickness, successful nitritation was demonstrated in a lab-scale MBBR process, achieving a nitrite accumulation ratio above 75% (Piculell et al., 2016a). Although no clear correlations could be made, it was hypothesized that NOB suppression in the nitritation stage was a long-term effect of FA and/or FNA inhibition during reject exposure.

Both AOB and NOB can be inhibited by FA and FNA, with NOB generally being more sensitive (Anthonisen et al., 1976), and by achieving FA or FNA at the appropriate level of exposure it may be possible to suppress only NOB. However, this level is not obvious, with many different results reported in literature (Blackburne et al., 2007; Chung et al., 2006; Vadivelu et al., 2007, 2006a, 2006b). In addition, the inhibitory effect of a certain bulk concentration of FA or FNA on NOB in a larger aggregate may differ from that in a pure culture of suspended cells. Different inhibitory effects have been observed in studies on activated sludge, biofilms and pure cultures (Hawkins et al., 2010; Kim et al., 2008; Kouba et al., 2014), and nitrifiers are potentially more sensitive to inhibition in smaller aggregates and thinner biofilms than in larger clusters – a theory which is yet to be experimentally evaluated.

Processes that suppress NOB by switching the feed between mainstream and reject are aiming for a lasting effect after exposure. However, most studies mentioned above have focused on the inhibitory effect during exposure. There is hence a need to evaluate the long-term inhibitory effect on AOB and NOB following exposure, when biomass is returned to a non-inhibiting environment.

In this study a fully nitrifying MBBR system, fed with mainstream wastewater, was temporally exposed to reject water from sludge treatment to evaluate this as a possible strategy for NOB inhibition and nitrite production at real conditions. Two different carrier types, with an approximate biofilm thickness of 400 and 50 μm, respectively, were compared in the study, to determine whether biofilm thickness had any influence on the effect. Biofilm structure and thickness were compared between the two carrier types by optical coherence tomography (OCT). Fluorescence in situ hybridization (FISH) was used together with confocal laser scanning microscopy (CLSM) to determine biofilm composition and internal stratification.

2. Material and methods

2.1. Pilot plant and carriers

The carriers used in this study were colonized by nitrifying biofilm in a 0.5 m^3 MBBR-pilot reactor, located at Sjölunda wastewater treatment plant (Malmö, Sweden). During operation, the pilot reactor was fed with effluent from a municipal high-rate activated-sludge (HRAS) plant, with low organic content to ensure the cultivation of fully nitrifying biomass on the carriers (for more data on pilot operation and performance, see supplementary material).

The pilot reactor contained a mixture of two different types of carriers, Z400 and Z50 (Fig. 1 and Table S1), at a total filling degree of approximately 30%. The Z-carriers differ from conventional MBBR carriers, as the biofilm grows on the outside of the carrier and not inside voids. An external grid on the carrier surface protects the biofilm from scouring as the carriers collide in the reactor, aiming to control biofilm thickness to the height of the grid walls (Piculell et al., 2016b). For Z400 and Z50, the grid walls were 400 and 50 μm respectively, enabling the development of different biofilm thickness in the same pilot reactor. The available area for biofilm growth was defined as the grid compartment bottom area, and differs in the two carriers (0.0013 and 0.0011 m^2/carrier in Z400 and Z50, respectively). After 233 days of pilot operation, carrier samples were removed and used in inhibition trials on a weekly basis, with the final sample being taken on day 261.

2.2. Inhibition trials

The inhibition trials aimed to expose the fully nitrifying biofilms from the pilot reactor to reject water at different loading rates, and to evaluate the effect on ammonium and nitrite oxidation after exposure. For each trial, 100 pieces of sample carriers, either Z400 or Z50, were removed from the pilot reactor and placed in 1 L lab-scale MBBR reactors (0.13 m^2/reactor and 0.11 m^2/reactor for Z400 and Z50, respectively). Each inhibition trial was initiated with 3–4 days of continuous operation at mainstream conditions, when the reactors were fed with the same mainstream HRAS effluent as the pilot plant (19–34 mgNH_4-N/L) at 20 °C, after which the feed was switched to municipal reject water (870–1010 mgNH_4-N/L) and the temperature was increased to 30 °C. After 1–2 days of reject exposure, the reactors were returned to mainstream conditions and operated for an additional 2–4 days. In total, 10 trials were performed for each carrier type.

During mainstream operation, the feeding rate was adjusted to a loading rate of 0.5–0.6 gTN/L,d, to ensure high substrate availability independent of inlet concentrations. During reject exposure the loading rate was varied between 0.2 and 1.2 gTN/L,d in the different trials, to achieve different reactor conditions and to determine the ideal reject exposure for nitritation (see Table 1). The duration and span in loading rate for the reject exposure was determined based on previous experiences in continuous lab trials (Piculell et al., 2016a). Aeration was kept similar (0.3–0.5 L/m) for both mainstream and reject operation, to ensure that reactor mixing did not vary between trials, and that DO concentrations remained relatively stable throughout the trials (5.8 ± 0.8 and 5.2 ± 0.9 mg/L during mainstream and reject operation, respectively). Temperatures were maintained stable at 20 °C and 30 °C, using thermostatic baths, while pH varied as a result of varying feeding rate, load and activity in the reactors (7.6 ± 0.2 and 8.1 ± 0.7 during mainstream and reject operation, respectively).

Fig. 1. Carries used in the study; Z400 with 400 μm grid wall height (left), and Z50 with 50 μm grid wall height (right).