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# Development of partial nitrification as a first step of nitrite shunt process in a Sequential Batch Reactor (SBR) using Ammonium Oxidizing Bacteria (AOB) controlled by mixing regime



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#### HIGHLIGHTS

- A Sequential Batch Reactor (SBR) was used to perform complete partial nitrification.
- A novel DO control strategy depending on using the mixing speed has been developed.
- The effect of DO, pH, alkalinity, FA and FNA acid have been evaluated.
- The SBR has been successfully running at nitrogen loading rate up to 1.2 kg/(m<sup>3</sup>.d).
- The SBR has achieved an ARE of 99% and a NAR of 93% at NLR of 1.2 kg/(m<sup>3</sup>.d).

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#### ABSTRACT

Shortcut biological nitrogen removal is a non-conventional way of removing nitrogen from wastewater using two processes either nitrite shunt or deammonification. In the nitrite shunt process, the ammonia oxidation step stops at the nitrite stage, which is known as partial nitrification, then nitrite is directly reduced to nitrogen gas. Effective partial nitrification could be achieved by accumulating Ammonia Oxidizing Bacteria (AOB) and inhibiting Nitrite Oxidizing Bacteria (NOB). In this research, a novel control strategy has been developed to control the DO using the variable mixing regime in a suspended growth system using a Sequential Batch Reactor (SBR) in order to achieve a stable ammonia removal efficiency (ARE) and nitrite accumulation rate (NAR) at a high nitrogen loading rate (NLR). The new controlled SBR system has been successfully running at NLR up to 1.2 kg/(m³.day) and achieved an ARE of 98.6 ± 2.8% and NAR of 93.0 ± 0.7%.

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### 1. Introduction

The excessive nitrogenous compounds withdrawn in water streams from the effluent of wastewater treatment plants causes numerous problems for the aquatic system as it leads to eutrophication causing the excessive growth of algae and increase in the oxygen depletion and poisons in the aquatic life. To avoid the aforementioned negative effects, reducing these compounds level has been a manner of great importance either by physical-chemical processes or biological processes. Due to its higher efficiency and lower cost over physical-chemical processes, Biological Nitrogen Removal (BNR) processes have been adopted widely. Conventional BNR processes comprise two main practices: nitrification

and denitrification. Nitrification is the aerobic biological conversion of ammonia to nitrate with oxygen as electron acceptor via a group of autotrophic bacteria through two steps involving Ammonia Oxidizing Bacteria (AOB) and Nitrite Oxidizing Bacteria (NOB), respectively. However, these two steps conventional BNR processes require 2 mol of oxygen to oxidize the ammonia to nitrate. Afterthought, the nitrate is reduced via heterotrophic bacteria to nitrite and nitrogen gas, which also requires organic matter during the denitrification stage.

Hence, conventional BNR processes require high oxygen and external carbon sources along with a slow growth of the autotrophic and heterotrophic bacteria. To overcome the aforementioned challenges and reduce the energy required for nitrogen removal as well as improve the nitrogen removal of side stream high ammonia waste streams, Shortcut Biological Nitrogen Removal (SBNR) has been developed. Based on the fact that nitrite is an intermediate compound in both nitrification and denitrifica-

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tion, SBNR relies on the direct conversion of nitrite produced in the first step of nitrification to atmospheric nitrogen instead of oxidizing it to nitrate then reducing the latter back to the former. Shortcut Biological Nitrogen Removal implies the reduction of oxygen consumption during the aerobic phase by 25% as a result of skipping the nitrite oxidation to nitrate and consequently reduces the total energy required by 60% (Peng and Zhu, 2006). Additionally, SBNR eliminates the use of external electron donor by 40%; resulting from skipping the nitrate reduction to nitrite; which makes it suitable for wastewater with low carbon to nitrogen ratio. Shortcut Biological Nitrogen Removal also results in a significant decrease of the sludge production in nitrification and denitrification processes by 35% and 55%, respectively (Peng and Zhu, 2006). The SBNR process comprises both nitrite shunt and deammonification processes. In the deammonification process, 50% of the ammonia is oxidized to nitrite subsequently the remaining ammonia is oxidized anaerobically to nitrogen gas using the nitrite produced as electron acceptor carried out by Anaerobic Ammonium Oxidizing (Anammox) bacteria. On the other hand, nitrite shunt stimulates the first step of nitrification (nitritation) and inhibits the second step of the oxidation of nitrite, which is known as partial nitrification, and then denitrifies nitrite directly to nitrogen gas as shown in Fig. 1. In the partial nitrification process, the ammonia is oxidized to hydroxylamine (NH<sub>2</sub>OH) catalyzed by the enzyme ammonia monooxygenase (AMO). This step requires one molecular oxygen and two extra electrons, while the second step consists of the further oxidation of hydroxylamine catalyzed by the hydroxylamine oxidoreductase (HAO) enzyme generating 4 electrons. Two of them are returned to ammonia monooxygenase to support the first step of nitritation (Hooper et al., 1997). Furthermore, the remaining two electrons serve for the cell synthesis. Through the electron transport, 1.65 electrons are passed to the terminal electron acceptor O<sub>2</sub> which is then reduced to form H<sub>2</sub>O, while the remaining 0.35 electrons are used for the reduction of NAD+ to NADH through 'reverse electron flow' which has been suggested to be performed by the embedded electron carrier NADH dehydrogenase (Whittaker et al., 2000).

Recently, partial nitrification has been adopted widely either for the nitrite shunt process or intermediate nitrite generation step for the Anammox process. However, the majority of the studies in the literature have targeted to achieve an effluent of NO<sub>2</sub>:NH<sub>4</sub> molar ratio of 1.31 suitable for subsequent Anammox process (Klapwijk and Stichting Toegepast Onderzoek Waterbeheer, 1996). However, fewer studies have targeted to reach a complete oxidation of ammonia to nitrite (full partial nitrification) as a first step for the nitrite shunt process (Table 1).

To achieve nitrite accumulation and selectively inhibit NOB, several strategies has been used including (i) maintaining low dissolved oxygen concentration, (ii) controlling free ammonia (FA) and free nitrous acid (FNA) concentrations through temperature/ pH, and (iii) reducing the hydraulic retention time (HRT). Performing partial nitrification through controlling the DO concentration in suspended growth system is based on the differences between the Monod saturation constant of oxygen for AOB (0.3 mg/L) and NOB (1.1 mg/L) indicating the higher affinity of oxygen for AOB over NOB (Wiesmann, 1994). Furthermore, AOB and NOB are sensitive to free ammonia (FA) and free nitrous acid (FNA) concentrations. The inhibition limit for NOB is 0.1-1.0 mg N/L, whereas 10-150 mg N/L of free ammonia is required to inhibit AOB (Anthonisen et al., 1976). Additionally, NOB is more sensitive to free nitrous acid compared to AOB. FNA concentration of 0.4-1.7 mg-N/L resulted in a 50% reduction in AOB activity, while low concentrations of 0.01-0.2 mg-N/L started to inhibit NOB (Zhou et al., 2011). Based on the fact that pH and temperature influence the equilibrium of FA and FNA, regulating these parameters can be crucial for achieving partial nitrification. Moreover, shortening HRT for AOB is an effective method to control the partial nitrification due to limited doubling time for AOB (7–8 h) compare to 50% more for NOB (Bock et al., 1986). The DO limitation is considered being the most feasible strategy for sustainable partial nitrification. However, due to the complexity of maintaining a uniform specific DO concentration, different strategies are required to maintain low DO concentrations and minimize the energy requirement during the SBNR. Therefore, 3 methods for aeration control during SBNR have been developed including (i) DO control using DO probe to control the Variable Frequency Drive (VFD) connected to the system blower, (ii) Ammonia Based Aeration Control (ABAC) using an ammonia probe to predict the required air flow rate according to the ammonia concentrations present in the system, and (iii) Ammonia vs. NO<sub>v</sub> (AvNTM) Control, which nitrifies only the amount of ammonia that can be denitrified afterwards. However, slow mixing speed accompanied with low aeration requirements could result in some biomass settling during the reaction time.

In this study, a novel strategy has been developed to control the DO to achieve a higher partial nitrification rate at a maximum NLR targeting complete ammonia oxidation to nitrite as a first step of

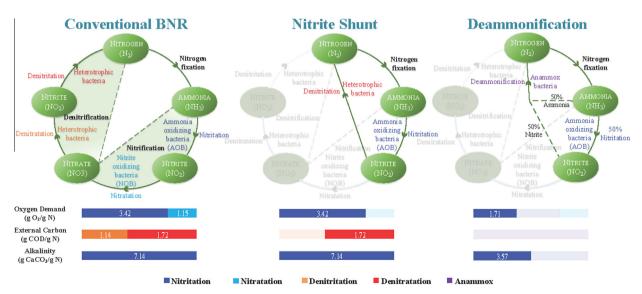


Fig. 1. Comparison between conventional and shortcut nitrogen removal over the nitrogen cycle.

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