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Advancing post-anoxic denitrification for biological nutrient removal

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ABSTRACT

The objective of this research was to advance a fundamental understanding of a unique post-anoxic denitrification process for achieving biological nutrient removal (BNR), with an emphasis on elucidating the impacts of surface oxygen transfer (SOT), variable process loadings, and bioreactor operational conditions on nitrogen and phosphorus removal. Two sequencing batch reactors (SBRs) were operated in an anaerobic/aerobic/anoxic mode for over 250 days and fed real municipal wastewater. One SBR was operated with a headspace open to the atmosphere, while the other had a covered liquid surface to prevent surface oxygen transfer. Process performance was assessed for mixed volatile fatty acid (VFA) and acetate-dominated substrate, as well as daily/seasonal variance in influent phosphorus and ammonia loadings. Results demonstrated that post-anoxic BNR can achieve near-complete (>99%) inorganic nitrogen and phosphorus removal, with soluble effluent concentrations less than 1.0 mgN L^{-1} and 0.14 mgP L^{-1}. Observed specific denitrification rates were in excess of typical endogenous values and exhibited a linear dependence on the glycogen concentration in the biomass. Preventing SOT improved nitrogen removal but had little impact on phosphorus removal under normal loading conditions. However, during periods of low influent ammonia, the covered reactor maintained phosphorus removal performance and showed a greater relative abundance of polyphosphate accumulating organisms (PAOs) as evidenced by quantitative real-time PCR (qPCR). While GAOs were detected in both reactors under all operational conditions, BNR performance was not adversely impacted. Finally, secondary phosphorus release during the post-anoxic period was minimal and only occurred if nitrate/nitrite were depleted post-anoxically.

1. Introduction

Phosphorus (P) and nitrogen (N) are nutrients of primary concern in regard to accelerated surface water eutrophication, and many wastewater treatment plants (WWTPs) are facing increasingly stringent effluent limitations for both nutrients. P and N can be readily removed biologically, with P removal achieved using an engineered process known as enhanced biological P removal (EBPR). EBPR is accomplished by exposing microbes to cyclical anaerobic/aerobic and/or anoxic conditions, with influent wastewater first directed to the anaerobic zone. The prescriptive EBPR configuration provides a selective advantage to organisms capable of storing volatile fatty acids (VFAs) anaerobically as polyhydroxyalkanoates (PHAs), such as polyphosphate accumulating organisms (PAOs), which remove and store excess P as intracellular polyphosphate (poly-P) and are the putative organisms responsible for EBPR. EBPR can also enrich for glycogen accumulating organisms

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(GAOs), which do not appear to contribute to EBPR and are therefore considered undesirable. PAOs generate energy for VFA uptake through hydrolysis of intracellular poly-P and glycogen degradation through glycolysis (Smolders et al., 1994), with glycogen degradation considered the main source of reducing power ($NADH_2$) for PHA storage (Zhou et al., 2010). Under aerobic and/or anoxic conditions, PAOs oxidize PHA via the TCA cycle to provide energy for growth, glycogen replenishment, P uptake, and poly-P storage (Smolders et al., 1995). As will be discussed later, *Candidatus* "Accumulibacter phosphatis" (henceforth referred to as Accumulibacter) has been suggested to be a dominant PAO, based on lab-scale and full-scale studies (He et al., 2007). GAOs exhibit similar metabolisms, with the exclusion of P cycling. Extensive research on factors affecting the PAO-GAO competition can be found elsewhere (Lopez-Vazquez et al., 2009; Oehmen et al., 2010a, 2007).

The combination of P and N removal is referred to as biological nutrient removal (BNR). Most BNR WWTPs accomplish denitrification using a pre-anoxic configuration, where the anoxic zone is located upstream of the aerobic zone. Since denitrification relies on ammonia oxidation in an aerobic zone, high mixed liquor recycle (MLR) rates are needed to provide a nitrate source in the anoxic zone. Although high specific denitrification rates (SDNRs) can be obtained with this configuration, there are several disadvantages associated with MLR pumping: higher energy costs, dissolved oxygen (DO) return from the aerobic, and dilution of influent carbon. Most importantly, the removal of oxidized nitrogen (NO_x; nitrate + nitrite) is ultimately limited by the MLR rate, and complete NO_x removable is unattainable (estimated 3–5 mg L^{-1} effluent total N) (Tchobanoglous et al., 2003).

Post-anoxic denitrification eliminates the need for MLR pumping, since the anoxic tank is located downstream of the aerobic nitrifying tank, and can produce effluent less than 3 mg L^{-1} total N (Tchobanoglous et al., 2003). In a non-EBPR system, an exogenous carbon source is typically supplied to drive denitrification. However, this approach cannot be applied to an EBPR system because the addition of carbon could promote phosphorus release (Kuba et al., 1994) and/or lead to proliferation of ordinary heterotrophic organisms which are incapable of excess P removal. Instead, a dual-sludge system is employed to separate the PAO and nitrifying sludges (Bortone et al., 1996; Kuba et al., 1996b). The PAO sludge bypasses nitrification, and intracellular PHA is thus conserved for post-anoxic denitrification. While the dual-sludge configuration eliminates the need for MLR pumping, it requires more underflow pumping and a larger footprint due to additional settlers.

An alternative post-anoxic EBPR-based configuration would leverage residual PAO carbon reserves (PHA and/or glycogen) to drive denitrification. In this operating scenario, use of the influent organic carbon and associated electrons is maximized for efficient nutrient removal. Further, this process configuration could produce lower effluent N and P loads as compared to traditional BNR configurations. Promising results have been obtained with lab- and pilot-scale continuous flow membrane bioreactors (Bracklow et al., 2010; Vocks et al., 2005) and lab-scale sequencing batch reactors (SBRs) (Coats et al., 2011b). These systems achieved SDNRs in excess of endogenous rates, and exhibited high N and P removal efficiencies. Recognizing the potential of this novel post-anoxic BNR process, and considering prior work, the research presented and discussed herein focused on understanding the effects of process operation and wastewater loading on N and P removal. New insight on relevant post-anoxic maintenance metabolisms is provided, and the issue of secondary P release is examined. This research also considered the long-term effects of surface oxygen transfer (SOT) on the anaerobic and anoxic aspects of the process, which has not been studied in relation to EBPR. Others have observed impaired SDNRs as result of SOT/microaerophilic conditions in open anoxic basins (Martins et al., 2004; Oh and Silverstein, 1999; Plósz et al., 2003), and therefore the issue could be especially relevant for a carbon-limited post-anoxic environment. The research also interrogated respective microbial consortia on PAO and GAO fraction. The research presents results based on the use of real municipal wastewater rather than the much more common approach of using synthetic wastewater.

2. Materials and methods

2.1. Experimental setup

Two independent SBRs (reactors O and C) were operated for over 250 days, with monitoring events as shown (Fig. 1). The reactors were operated identically except for the headspace condition; reactor O had a headspace open to the atmosphere, while reactor C had zero headspace due to a liquid surface covered with a polyethylene disk. Thus, SOT could occur during the anaerobic and anoxic periods in reactor O but not in reactor C. Note that for all of the figures in this manuscript, reactor O is represented by open symbols, while reactor C is denoted by filled symbols. Each SBR (0.9 L operating volume) was inoculated with activated sludge obtained from the Moscow, ID WWTP, which operates a hybrid A^2/O-oxidation

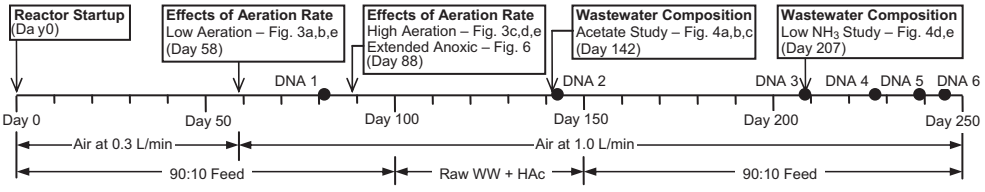

Fig. 1 – Research timeline showing time points for all sampling investigations in this study (aeration rate and substrate are provided below the timeline).