

Feasibility of Focused–Pulsed Treated Waste Activated Sludge as a Supplemental Electron Donor for Denitrification

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ABSTRACT: We evaluated the feasibility of using waste activated sludge (WAS) from a wastewater treatment plant as an internal electron donor to fuel denitrification, by increasing its bioavailability with Focused–Pulsed (FP) technology. The focused–pulsed treatment of WAS (producing FP-WAS) increased the semi-soluble chemical oxygen demand (SSCOD) by 26 times compared with the control WAS. The maximum denitrification rate of FP-WAS (0.25 g nitrate-nitrogen [NO₃⁻-N]/g volatile suspended solids [VSS]·d) was greater than for untreated WAS (0.05 g NO₃⁻-N/g VSS·d) and methanol (0.15 NO₃⁻-N/g VSS·d). Centrifuging out the larger suspended solids created FP-centrate, which had a rate (0.14 g NO₃⁻-N/g VSS·d) comparable with that of methanol. Thus, FP treatment of WAS created SSCOD, which was an internal electron donor that was able to drive denitrification at a rate similar to or greater than methanol. One trade-off of using FP-WAS for denitrification is an increase in total Kjeldahl nitrogen (TKN) loading. While FP-WAS achieved the lowest total nitrogen and NO₃⁻-N concentrations in the batch denitrification test, its final ammonia-nitrogen (NH₃-N) concentration was the highest, as a result of the release of organic nitrogen from the FP-treated biomass; FP-centrate had less release of total soluble nitrogen. While the return of total nitrogen (TN) is small compared with the SSCOD, the effects of the added nitrogen loading need to be considered. *Water Environ. Res.*, **82**, 2316 (2010).

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Introduction

A variety of pre-denitrification processes have been developed as modifications of the activated sludge process (e.g., anaerobic-anoxic-oxic [A²/O] [Sedlak, 1991], Modified Ludzack–Ettinger [MLE] [Ludzack and Ettinger, 1962], and 4-stage Bardenpho [Barnard, 1975]). In these processes, the total nitrogen removal efficiency depends on the internal recycle ratio from an aerobic zone to an anoxic zone and the chemical oxygen demand (COD)-to-nitrogen ratio in the influent wastewater. Practical limits on the recycle ratio make achieving very high total nitrogen (TN) removals impossible in pre-denitrification processes, and a low

COD/N ratio exacerbates the problem. Thus, addition of a supplementary electron donor and multi-stage denitrification are necessary to achieve advanced removal of nitrogen.

Since the work of McCarty et al. (1969), methanol has been the most common external electron donor for heterotrophic denitrification, because it is relatively inexpensive. Despite its widespread use, methanol gives slower denitrification kinetics than fumarate, ethanol, formate, acetate, butyrate, valerate, and propionate (Oa et al., 2006; Xu, 1996), probably because it must be oxidized to formate or formaldehyde before denitrifiers can metabolize it (Constantin and Fick, 1997; Xu, 1996). In addition, methanol is acutely toxic to humans and presents severe handling difficulties. Moreover, the price of methanol changes with the price of fossil fuels (Olah et al., 2006). Thus, finding good alternatives to methanol is a top priority in wastewater treatment facilities that use it as an external electron donor. Substituting another simple organic compound, such as acetate, is technically feasible, but typically is much more costly (Tecnon OribiChem, 2010).

A typical wastewater treatment plant (WWTP) has a natural internal electron donor in its waste activated sludge (WAS). Therefore, reusing the WAS as an electron donor for denitrification might replace the addition of external sources. However, WAS, already being well-stabilized, has a low fraction of readily bioavailable COD. Hence, a pretreatment technique is needed to increase the bioavailability of WAS.

So far, several thermal, physical, and chemical sludge treatment technologies have been developed to increase the bioavailable COD (Eskicioglu et al., 2007). For example, the soluble COD (SCOD) was increased by 361 ± 45% and 143 ± 34% by thermal treatment of WAS (96°C) and microwave irradiation (1250 W, 2450 MHz), respectively (Eskicioglu et al., 2006). An increase of the SCOD-to total COD (TCOD) ratio from 0.05 (control) to 0.2 was reported for ultrasound pretreatment of municipal WAS (20 kHz, 110 W for 120 minutes [Chu et al., 2001]).

Focused–pulsed (FP) treatment is a new entry for sludge pretreatment and has demonstrated significant benefits at full-scale when used to enhance anaerobic digestion to produce methane (CH₄) (Rittmann et al., 2008; Salerno et al., 2009; Zhang et al., 2009). FP treatment ruptures the cell membrane and cell wall of cells by exposing them to a series of short, high-voltage pulses (20 to 30 kV at 2000 to 3000 Hz). Even though the voltage is high, the treatment time is very short (less than 1 second). FP treatment expands existing pores (electroporation), creates new pores in the cell membrane, and breaks the cell into fragments (Ohio State University, 2010; Salerno et al., 2009). FP treatment increases

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SCOD from WAS, but the bioavailable COD increases much more (Salerno et al., 2009). The largest part of the bioavailable COD is comprised of small colloids, which are called *semi-soluble COD* (SSCOD). An important question concerns whether the SSCOD is used rapidly enough to drive denitrification for advanced total nitrogen removal. This aspect can be judged by its kinetics in comparison with alternative organic donors, including pure organic chemicals (e.g., methanol and acetate) and organic waste streams.

One side product of using treated WAS as the electron donor in denitrification is that hydrolysis of the protein fraction releases ammonia-nitrogen (NH₃-N) and organic nitrogen in proportion to bioavailable COD. For example, soluble protein increased from 88 to 224 mg/L and from 88 to 124 mg/L by microwave and heat treatment of WAS, respectively (Eskicioglu et al., 2007). In research with mechanical treatment, increases were 250% for soluble protein and 20% for NH₃-N (Nah et al., 2000), while soluble nitrogen increased by 120 to 200% after pulsed-power treatment (Choi et al., 2006). The effectiveness of treated WAS as an electron source is diminished if the released NH₃ is too great, because released NH₃ counteracts the goal of total nitrogen removal. Therefore, the increase of nitrogen in relation to bioavailable COD must be evaluated to use pretreated WAS as a denitrification electron donor. Likewise, the increase of soluble phosphorus after FP treatment is also a potential concern and should be evaluated.

Another side effect of using WAS as the donor for denitrification is that it returns volatile suspended solids (VSS) to the wastewater treatment train. In a WWTP, centrate or supernatant typically is produced from sludge thickening before anaerobic digestion and returned to the headworks to be treated. If the treated WAS were thickened so that the centrate could be used directly as the electron donor for denitrification, the loadings of solids and poorly bioavailable COD could be minimized. Therefore, the centrate of pretreated WAS may be an advantageous electron source to avoid solids buildup.

Another aspect of using WAS to fuel denitrification is that it diverts COD from methane generation at the same time that it cuts down the chemical expense for the external electron donor (e.g., methanol) required for denitrification. Based on current prices for methanol and methane (methanol = \$0.20/L CH₃OH [Methanex Corporation, 2010] and CH₄ = \$0.17/m³ CH₄, Energy Information Administration, 2010), the calculations below show that the savings from not using methanol are greater than the value of the lost CH₄—\$0.16/kg COD-methanol versus \$0.12/kg COD-CH₄.

Unit cost of methanol:

$$\begin{aligned} & \$0.20/\text{L CH}_3\text{OH} \times 1 \text{ L CH}_3\text{OH}/0.791 \text{ kg CH}_3\text{OH} \times \\ & 32 \text{ kg CH}_3\text{OH} / 6000 \text{ e}^- \times 1000 \text{ ke}^- / 8 \text{ kg COD} = \\ & \$0.16/\text{kg COD} \end{aligned}$$

Unit cost of CH₄:

$$\begin{aligned} & \$0.17/\text{m}^3 \text{ CH}_4 \times 22.4 \text{ m}^3 \text{ CH}_4 / 1 \text{ kmol CH}_4 \times 1 \text{ kmol} \\ & \text{CH}_4 / 4000 \text{ e}^- \times 1000 \text{ e}^- / 8 \text{ kg COD} = \$0.12/\text{kg COD} \end{aligned}$$

We applied FP treatment to WAS (called *FP-WAS*) to evaluate its ability to increase bioavailable COD as an electron donor for denitrification. We also centrifuged the FP-WAS to separate liquid and solids; we call this liquid fraction *FP-centrate*. First, we compared the properties of FP-WAS and FP-centrate with the untreated WAS to assess the effect of focused-pulsed treatment. Second, we performed batch denitrification tests to compare the

denitrification kinetics and the COD availability among acetate, methanol, FP-WAS, and FP-centrate. Finally, we evaluated the effects of recycling NH₃-N, phosphorus, and suspended solids.

Materials and Methods

Waste Activated Sludge and Focused-Pulsed Treatment. We obtained WAS from the secondary clarifier underflow at the Mesa Northwest Water Reclamation Plant (MNWWRP) in Mesa, Arizona, which uses the anoxic-oxic (A/O) process. At the time we obtained WAS, methanol was added to the headworks (before the primary settling tank) as the external electron donor to remove nitrogen by denitrification.

We FP-treated the WAS to increase the bioavailable COD for use as a denitrification electron donor. Details of the FP treatment system are presented in Salerno et al. (2009). Several variables of FP treatment can affect the increase of bioavailable COD—applied voltage, pulse width, frequency, and hydraulic residence time in the treatment chamber. To normalize the intensity of FP treatment, we use the treatment intensity (Salerno et al., 2009), which combines the key parameters of focused-pulsed treatment.

$$\text{Treatment Intensity} = K \cdot \frac{V^2 \cdot D \cdot f \cdot \sigma \cdot \text{HRT}}{L^2} \quad [\text{kWh}/\text{m}^3] \quad (1)$$

Where

- V = applied voltage ($V = J/C = \text{kg} \cdot \text{m}^2/\text{C} \cdot \text{s}^2$),
- D = pulse width (sec/pulse),
- f = pulse frequency (pulse/sec),
- σ = sample conductivity ($\text{S}/\text{m} = \text{s} \cdot \text{C}^2/\text{kg} \cdot \text{m}^3$),
- L = distance between electrodes (m),
- HRT = residence time in the pulsed electric field (seconds), and
- K = a constant for unit conversion (1/3 600 000 kWh/J).

We carried out FP treatment of primary sludge plus WAS with a treatment intensity of 16 kWh/m³ before full-scale methanogenic digestion, in which the CH₄ production rate was increased by approximately 30% when approximately 63% of the input sludge was FP pre-treated (Rittmann et al., 2008; Zhang et al., 2009). Here, we used a treatment intensity of 28 kWh/m³ of sludge treated by FP treatment, which was 1.7 times higher than the treatment intensity of 16 kWh/m³. Assuming that 380 m³ of sludge is produced from 76 000 m³ of wastewater treatment, 28 kWh/m³ of energy consumption by FP treatment on sludge can be converted to 0.14 kWh/m³-wastewater, which compares with a total energy requirement of 1.22 kWh/m³-wastewater for air-activated sludge with nitrification and denitrification (Pacific Gas and Electric Company, 2003). Thus, the energy input for FP treatment of all the waste sludge is equal to approximately 10% of the total energy consumption for treatment before FP treatment.

Effect of Focused-Pulsed Treatment on Waste Activated Sludge Properties. We evaluated how focused-pulsed treatment and centrifugation affected COD, nitrogen, and suspended solids in WAS, as these are key factors to determine the feasibility of using FP-treated sludge or its centrate as the electron donor for denitrification. We analyzed total COD (TCOD), total suspended solids (TSS), Volatile suspended solids (VSS), and semi-soluble parameters—COD, NH₃-N, TN, total phosphorus (TP), proteins, sugars, and volatile fatty acids (VFAs). The semi-soluble samples, which contained small colloids and soluble materials, were

Table 1—Batch-experiment conditions for testing denitrification kinetics and stoichiometry. The conditions were the same for the two batch tests.

Electron donor	Inoculum ^a	Inoculum VSS (mg/L)	SSCOD (mg/L)	NO ₃ ⁻ -N (mg/L)
Control	0.5 L	2770	16.7 ^b	62.4
Acetate	0.5 L	2770	86.7	50.9
Methanol	0.5 L	2770	110.5	55.5
Untreated-WAS	0.2 L ^c	2770 (5650) ^d	29.7	61.2
FP-WAS	0.2 L ^c	2770 (5350) ^d	120.5	60.0
FP-centrate ^e	0.2 L ^c	2770 (2920) ^d	115.2	59.7

^a Anoxic mixed-liquor suspended solids (MLSS) taken from anoxic tank in MNWWRP.

^b Originated from the anoxic inoculums.

^c Volume after settling to obtain the same amount of anoxic inoculum as the control, acetate, and methanol cases.

^d Total VSS in the denitrification test bottle resulting from the addition of VSS with the electron donor.

^e Centrate after focused-pulsed treatment and centrifugation at 2000 rpm for 10 minutes.

prepared by filtering the samples through a 1.2- μ m glass-fiber filter (Whatman GF-C, Maidstone, United Kingdom).

We measured COD using a Hach COD kit (concentration range = 10 to 1500 mg/L) (Hach Company, Loveland, Colorado), TN using a Hach T-N kit (concentration range = 10 to 150 mg/L), NH₃-N using a Hach NH₃-N kit (concentration range = 0 to 50 mg/L as nitrogen), and VFAs using a Hach volatile acids reagent set. The wavelengths to measure COD, TN, semi-soluble-NH₃-N, and semi-soluble VFAs in a spectrophotometer (Genesys 20, Thermo Spectronic, Altham, Massachusetts) were 620, 410, 655, and 495 nm, respectively. Organic nitrogen was calculated by subtracting NH₃-N from TN. We measured TP using a Hach phosphorous kit (UHR TNT 845, 2 to 20 mg/L PO₄-P) and Hach DR-2800 spectrometer.

Sugars and proteins were analyzed by a colorimetric method (Bubois et al., 1956) and the bicinchoninic acid method (Brown et al., 1989), respectively. For these analyses, we developed standard calibration curves with glucose and bovine serum albumin, respectively. The TSS and VSS were determined by weight according to *Standard Methods* (APHA et al., 1998).

Denitrification Batch Tests Using Different External Electron Donors. The FP-WAS, FP-centrate, untreated WAS, methanol, and acetate were compared directly, in terms of denitrification rate and stoichiometry using batch tests. We obtained the inoculum for the batch experiments from one of the two parallel anoxic tanks at the MNWWRP, where methanol was used as the external electron donor for denitrification. For the denitrification tests, we used 1-L Erlenmeyer flasks shaken at 170 rpm and held at a temperature of 25°C. After inoculation, we continually purged the flasks with nitrogen gas (N₂) to remove and exclude oxygen and then added nitrate-nitrogen (NO₃⁻-N) and the selected electron donor to each flask, which defined the start of the batch test. The target COD/N ratio for initial electron donor and electron acceptor concentrations was 2 g COD:g N, which is lower than stoichiometry for complete denitrification (3.70 g COD/g N). The reason for using the lower COD/N ratio was to avoid electron-acceptor limitation, so that the kinetics for the donor could be evaluated directly. Based on this ratio, approximately 60 mg/L NO₃⁻-N and approximately 120 mg SSCOD/L of acetate, methanol, FP-WAS, or FP-centrate were added as electron acceptor and electron donor, respectively. We also prepared a control having no external electron donor and an untreated WAS control, each having the same volume of untreated

WAS as for the tests with FP-WAS and FP-centrate as electron donor. In the case of FP-WAS, FP-centrate, and untreated-WAS, we concentrated 0.2 L of anoxic inoculum by settling for approximately 1 minute and then seeded the settled solids to the microcosms to maintain the same amount of mixed-liquor volatile suspended solids (MLVSS) concentration as the other cases. For acetate and methanol, we did not concentrate the inoculum solids. The quantities of the various electron donors, electron acceptor (NO₃⁻-N), and anoxic inoculum used for batch tests were averaged and are listed in Table 1. We duplicated the batch experiments, which gave similar results, and we report the average data for all parameters.

The total duration of a denitrification batch experiment was 3 hours, and we measured semi-soluble COD, NH₃-N, NO₂⁻-N, and NO₃⁻-N over time for each case. The NO₂⁻-N and NO₃⁻-N were analyzed using an ion chromatograph (ICS-3000, Dionex, Sunnyvale, California) equipped with an AS-18 Guard, Ionpac AS-18 anion exchange column (Dionex) and conductivity detector after filtering with a 0.2- μ m polyvinylidene fluoride (PVDF) membrane filter (Pall Life Sciences, Ann Arbor, Michigan). Potassium hydroxide (KOH) was used as an eluant (22 mM for 0 to 9 minutes and 35 mM for 9 to 35 minutes).

Results and Discussion

Properties of Focused-Pulsed-Treated Waste Activated Sludge. Table 2 summarizes the properties of WAS before and after FP treatment. The TSS and VSS of the WAS decreased by 480 and 270 mg/L, respectively, after FP treatment, meaning that a portion of suspended solids was solubilized or fragmented to small colloids. After FP treatment, SSCOD, sugars, proteins, and VFAs increased 26-, 4.9-, 5.0-, and 5.1-fold, respectively, compared with the untreated WAS. The increase of these parameters most likely was related to membrane breakage and cell fragmentation resulting from FP treatment rather than to an increase in temperature, because the temperature increase after FP treatment was only approximately 17.7(\pm 3)°C. After FP treatment, the SSCOD/TCOD ratio, which is a general indicator of the extent of hydrolysis and breakage, increased from 0.15% in the untreated WAS to 4.0% for FP-WAS.

The last column in Table 2 provides the same values for FP-centrate. As expected, centrifugation of FP-WAS removed all the suspended solids. The concentrations for the semi-soluble components were slightly higher in the FP-centrate than FP-WAS, which suggests that centrifugation may have enhanced

Table 2—Characteristics of WAS after Focused-Pulsed treatment.

Parameter	Concentration (mg/L)		
	Untreated-WAS	FP-WAS	FP-centrate ^a
TSS	8480 (± 1770) ^b	8000 (± 1710)	-
VSS	6360 (± 1290)	6090 (± 1270)	-
TCOD	8510 (± 2120)	8430 (± 1990)	ND ^c
Semi-soluble			
COD	12.8 (± 6.0)	336 (± 124)	397 (± 152)
NH ₃ -N	7.0 (± 4.0)	13.6 (± 5.2)	15.7 (± 3.5)
TN	7.3 (± 4.0)	25.3 (± 10.1)	27.0 (± 9.0)
TP	3.4 (± 0.1)	22.2 (± 0.4)	-
Protein	33 (± 3.6)	167 (± 40.2)	187 (± 38.3)
Sugar	4.9 (± 0.9)	24.0 (± 8.5)	31.4 (± 10.1)
VFAs	4.9 (± 1.5)	24.9 (± 11.2)	27.9 (± 10.5)

^a We prepared FP-centrate sample by centrifuging FP-WAS at 2000 rpm for 10 minutes.

^b Numbers in parentheses are standard deviations for treatment of different batches of WAS; the range of %RSD (relative standard deviation = measure the precision of the analysis = (average/standard deviation)^{1/2} × 100) for MLSS, MLVSS, and TCOD was 21–25% ($n = 4$), but for semi-soluble properties was 11 to 57% (untreated-WAS) and 24 to 45% (FP-WAS), which means the focused-pulsed treatment efficiency can be varied by raw WAS properties.

^c ND= Not determined.

biomass disruption. However, the more important trend is that the semi-soluble concentrations were similar between FP-WAS and FP-centrate, which means that filtration through a 1.2- μ m glass fiber filter provided a good measure of the semi-solid characteristics after centrifugation.

Increased bioavailable COD and semi-soluble total nitrogen (SSTN) are the critical parameters to determine whether FP-treated materials can be used efficiently as an electron donor for denitrification in a WWTP. The ideal situation is that the bioavailable COD increases by a large amount, because it is the donor, but that semi-soluble total nitrogen does not increase much, because increased total nitrogen adds to the amount of nitrogen that must be nitrified and eventually denitrified. FP treatment increased the SSCOD from 12.8 mg/L (untreated-WAS) to 336 mg/L (FP-WAS) and 397 mg/L (FP-centrate), while SSTN increased from 7.3 to 25.3 mg/L (FP-WAS) and 27.0 mg/L (FP-centrate) by FP treatment. Comparing the FP-centrate to the untreated-WAS, the extent of SSCOD increase (31.0 times) was much higher than the increase in SSTN (3.7 times). The stoichiometric ratio of grams of COD needed to denitrify 1 gram of nitrogen is 3 to 4 g COD/g N (Rittmann and McCarty, 2001). The increase as SSCOD/SSTN was 31.0/3.7 = 8.4 g COD/g N, which is approximately 2.5 times the stoichiometric demand. In terms of absolute concentration, the ratio of SSCOD/SSTN in the FP-centrate was 397/27 = 14.7 g COD/g N, which is favorable for driving denitrification without adding excessive nitrogen.

FP treatment also increased the concentrations of all of the other semi-soluble components. Perhaps most notable is the increase in total phosphorus from 3.4 to 22.2 mg/L, because total phosphorus can be regulated in the effluent, but cannot be oxidized biologically if it is returned with the FP-WAS or FP-centrate.

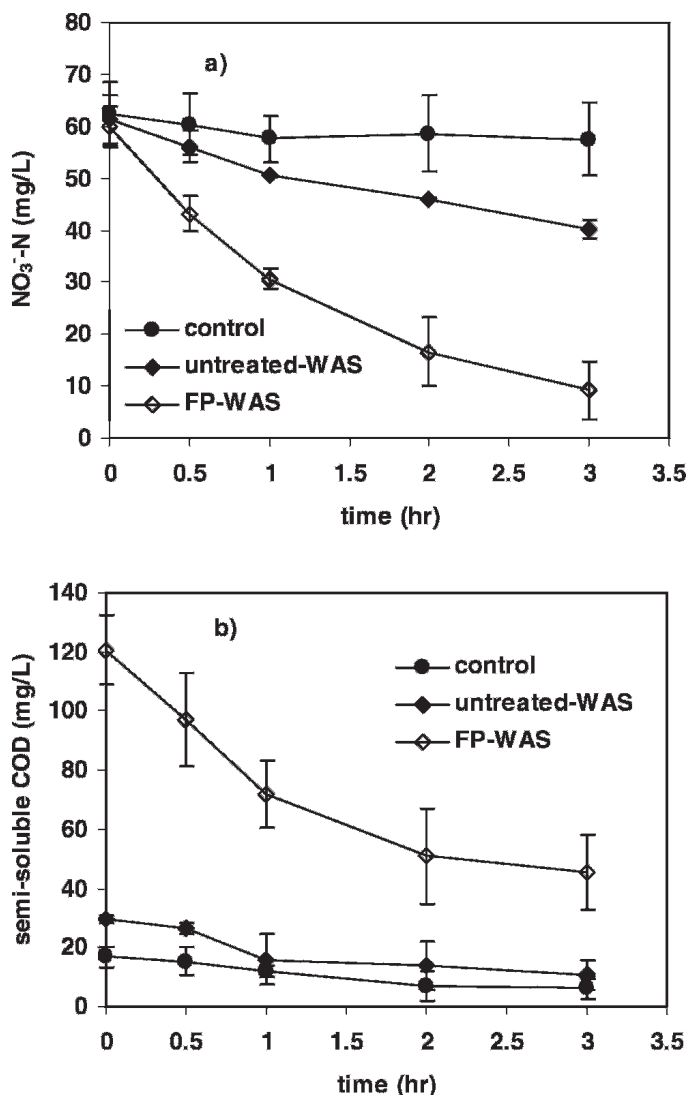


Figure 1—Comparison of NO₃⁻-N and SSCOD reductions with untreated-WAS and FP-WAS as an electron donor and with a no-donor control: (a) NO₃⁻-N and (b) SSCOD. NO₂⁻-N was not detected. The conditions were the same for the two batch tests, and the data presented are the average (symbol) with range (bars).

Comparing Denitrification Kinetics for Untreated Waste Activated Sludge and Focused-Pulsed-Treated Waste Activated Sludge as an Electron Donor. Figure 1 compares the denitrification kinetics for untreated WAS, FP-WAS, and a no-donor control. The NO₃⁻-N loadings at the beginning of the batch tests for untreated-WAS, FP-WAS, and no-donor control were 0.022, 0.022, and 0.023 mg NO₃⁻-N/g VSS, respectively, considering only the inoculum VSS in the denominator. Because the untreated WAS increased the mass of active biomass, its NO₃⁻-N loading was 0.011 mg NO₃⁻-N/g VSS considering all of the biomass.

FP treatment of WAS improved denitrification efficiency and absolute rate by more than 2 times compared with untreated WAS (Figure 1a). The denitrification efficiency increased from 34% for untreated-WAS to 85% for FP-WAS over the full 3-hour test, and it was only approximately 8% with no added donor. Likewise, the maximum specific denitrification rate (in the first hour) increased from 0.05 to 0.25 g NO₃⁻-N/g VSS·d based on initial active VSS in the batch bottles. The higher denitrification efficiency and rate

Table 3—Stoichiometries of SSCOD and nitrogen removals of the different materials tested as electron donors in batch denitrification experiments.

Electron donor	Removed SSCOD (mg)	Removed NO_3^- -N (mg)	$\Delta\text{SSCOD}/\Delta\text{N}$
Control	5.3	2.4	2.2
Untreated-WAS	9.5	10	0.9
FP-WAS	37	25	1.5
Acetate	30	13	2.3
Methanol	28	25	1.1
FP-centrate	37	16	2.3

for FP-WAS provide direct evidence for pre-treatment making the COD more bioavailable, because the same amount of WAS was added in each experiment. One reason for the difference is illustrated in Figure 1b, in which the starting SSCOD value was 121 mg/L for FP-WAS, compared with only 30 mg/L for untreated WAS.

The stoichiometry of nitrogen and COD removals is one of the important factors to evaluate the feasibility of any type of organic matter as an alternative electron donor for denitrification. The top part of Table 3 shows the relationships of removed SSCOD to removed NO_3^- -N in untreated WAS and FP-WAS during the denitrification tests. For comparison, the theoretical values with biomass growth for acetate and methanol are 4.85 g COD/g NO_3^- -N and 3.7 g COD/g NO_3^- -N, respectively. The stoichiometries of removed SSCOD to nitrogen in no-donor control, untreated-WAS, and FP-WAS for 3-hour denitrification tests were 2.2, 0.9, and 1.5 g COD/g NO_3^- -N, respectively, which are lower than the typical COD/N ratio for denitrification (approximately 4 g COD/g NO_3^- -N [Rittmann and McCarty, 2001]). The most likely reason for lower stoichiometry is that the high VSS (2770 to 5650 mg/L) in the denitrification batch reactors provided suspended-solids electron donor, which augmented the SSCOD. A less likely possibility for these tests is that the release of soluble microbial products increased the final SSCOD; this possibility is discussed further in the next section.

Comparing Denitrification Kinetics of Focused-Pulsed Treated Waste Activated Sludge Centrate with Methanol and Acetate as an Electron Donor. Figure 2 compares the denitrification kinetics of FP-centrate with the kinetics of methanol and acetate. The NO_3^- -N loadings at the beginning of batch tests with acetate, methanol, and FP-centrate were 0.018, 0.020, and 0.020 g NO_3^- -N/g VSS, respectively.

Denitrification started without lag in all cases. Acetate gave the fastest initial rate, while FP-centrate was similar to methanol. For the full 3-hour denitrification period, the nitrate-removal efficiencies for each electron donor were 66% (acetate), 92% (methanol), and 55% (FP-centrate). The reason for the relatively low denitrification efficiency with acetate might be its lower initial SSCOD compared with the other cases (Figure 2b). The lower SSCOD concentration with acetate, with a lower nitrate concentration (Figure 2a), may have been the result of denitrification in the sample during the approximately 10-minute time period before it could be filtered.

In parallel with the nitrate removals, SSCOD removals (Figure 2b) for methanol were lower than for FP-centrate, even though methanol gave greater total removed NO_3^- -N than FP-centrate. The final SSCOD concentrations in the methanol and FP-

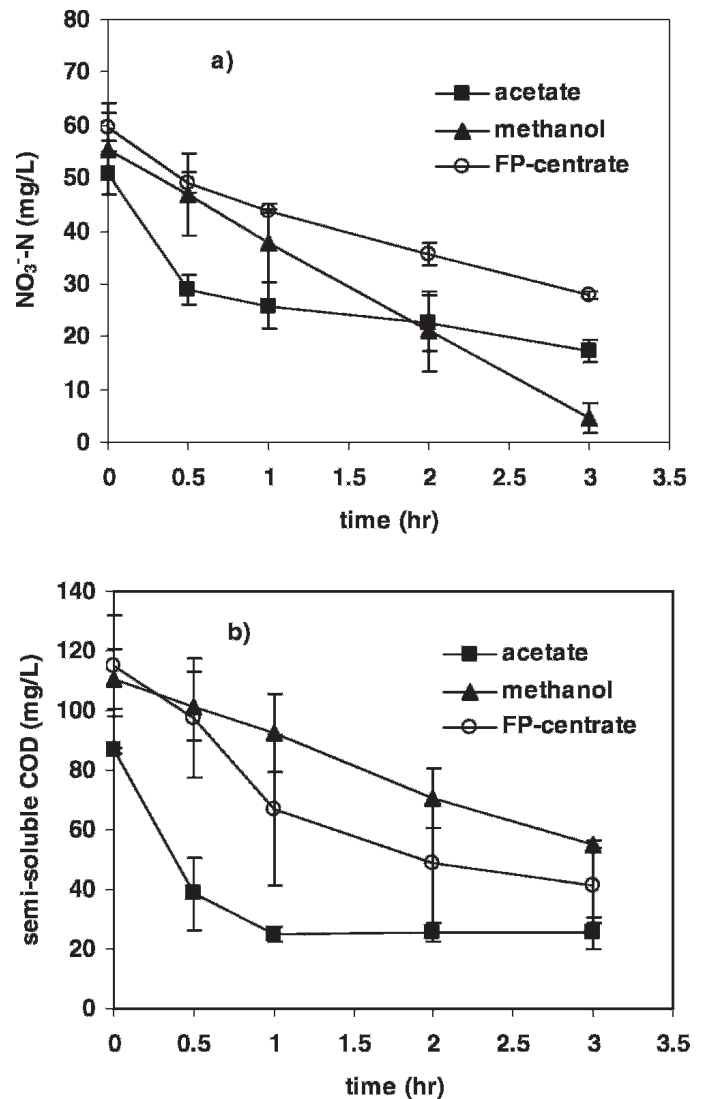


Figure 2—Time course of NO_3^- -N and SSCOD reductions with acetate, methanol, and FP-centrate as electron donor: (a) NO_3^- -N and (b) SSCOD. NO_2^- -N was not detected. The conditions were the same for the two batch tests, and the data presented are the average (symbol) with range (bars).

centrate cases were 55 and 41 mg/L, respectively. Based on these SSCOD removals, greater than 64% of the semi-soluble organic matter in FP-WAS and FP-centrate was bioavailable and could be used to drive denitrification. While pure chemicals are 100% bioavailable, 60% of COD in mecellulose wasted liquor and 65% of organic matter in thermal hydrolysate were usable for denitrification (AESoy et al., 1998; Park et al., 2005). The final SSCOD concentration of acetate, which is known to be a rapidly biodegradable substrate, was approximately 25 mg/L. The final SSCOD probably was from soluble microbial products (SMPs) from substrate metabolism and biomass decay. The SMPs have been found to account for the majority of soluble organic material in the effluent from biological treatment processes (de Silva and Rittmann, 2000; Rittmann et al., 1987; Schiener et al., 1998).

The bottom part of Table 3 shows the relationships of removed SSCOD and NO_3^- -N for acetate, methanol, and FP-centrate during the 3-hour denitrification tests. Consumed SSCOD for denitrification—2.3, 1.1, and 2.3 g/g N for acetate, methanol, and FP-centrate,

Table 4—Denitrification rates and semi-soluble COD removal rates using different electron donors.*

Electron donor	Maximum SSCOD removal rate (g SSCOD/g VSS·d)	Maximum denitrification rate (g N/g VSS·d)
Control	0.04	0.04
Untreated-WAS	0.06	0.05
FP-WAS	0.42	0.25
Acetate	0.83	0.33
Methanol	0.17	0.15
FP-centrate	0.42	0.14

* We considered only the VSS of the anoxic inoculum for calculating the specific rates in all cases except untreated-WAS, which provided additional active inoculum.

respectively—were lower than the typical COD/N ratio for denitrification (approximately 4 g COD/g NO_3^- -N). Elefsiniotis et al. (2004) also reported a COD/N ratio of 0.68 to 0.98, with an initial COD/N ratio of 0.4 to 1.8 in denitrification tests using VFAs as electron donors. The low SSCOD/N ratio suggests that the inoculum was providing endogenous donor or that SMPs were being released to increase the SSCOD. Because the no-donor control did not show the same kind of low ratio (first entry in Table 3), endogenous donor is less likely than release of SMP in these experiments, which had no addition of FP-treated VSS.

Table 4 gives the average and maximum specific denitrification rates for each case. Important trends from the rate data are the following:

(1) The denitrification rates using acetate and methanol were similar to the denitrification rates in other batch studies with acetate and methanol—for example, 0.29 g NO_3^- -N/g VSS·d

for acetate (Carrera et al., 2003) and 0.077 (Peng et al., 2007), 0.17 (Carrera et al., 2003), and 0.29 g NO_3^- -N/g VSS·d (Lee et al., 1995) for methanol.

(2) The denitrification rate with untreated-WAS was approximately double the rate of the control, which means that added VSS can be used to drive denitrification—0.09 and 0.04 g NO_3^- -N/g VSS·d. Both are similar to the rate of 0.084 g NO_3^- -N/g VSS·d reported for endogenous denitrification (Elefsiniotis et al., 2004).

(3) Acetate gave the highest denitrification rate, FP-WAS was second fastest, and FP-centrate or methanol was the third fastest electron donor.

Nitrogen Transformations. Figure 3 compares initial and final NO_3^- -N, NH_3 -N, and organic nitrogen concentrations in the semi-soluble phase among the WAS-based electron donors during the denitrification tests. Addition of untreated WAS had minimal effect on the initial SSTN, but addition of FP-WAS clearly increased the initial semi-soluble organic nitrogen and NH_3 -N, because both were released as part of the bioavailable organic matter during FP treatment. Addition of FP-centrate gave a similar increase of NH_3 -N, but a smaller increase of organic nitrogen. It is possible that the higher organic nitrogen content with FP-WAS was caused by release from incompletely damaged cells during the time that the FP-centrate was being prepared by centrifugation.

Except for the no-donor control, the final SSTN concentrations were reduced after the denitrification test. The large nitrogen losses were from denitrification of NO_3^- -N. The lowest final concentration of NO_3^- -N was with FP-WAS, and this also allowed the lowest final TN. Untreated WAS, FP-WAS, and FP-centrate showed increases in NH_3 -N of 0.8, 8.2, and 4.5 mg/L, respectively, as a result of hydrolysis of organic nitrogen during the denitrification tests; FP-WAS had the largest increase in NH_3 -

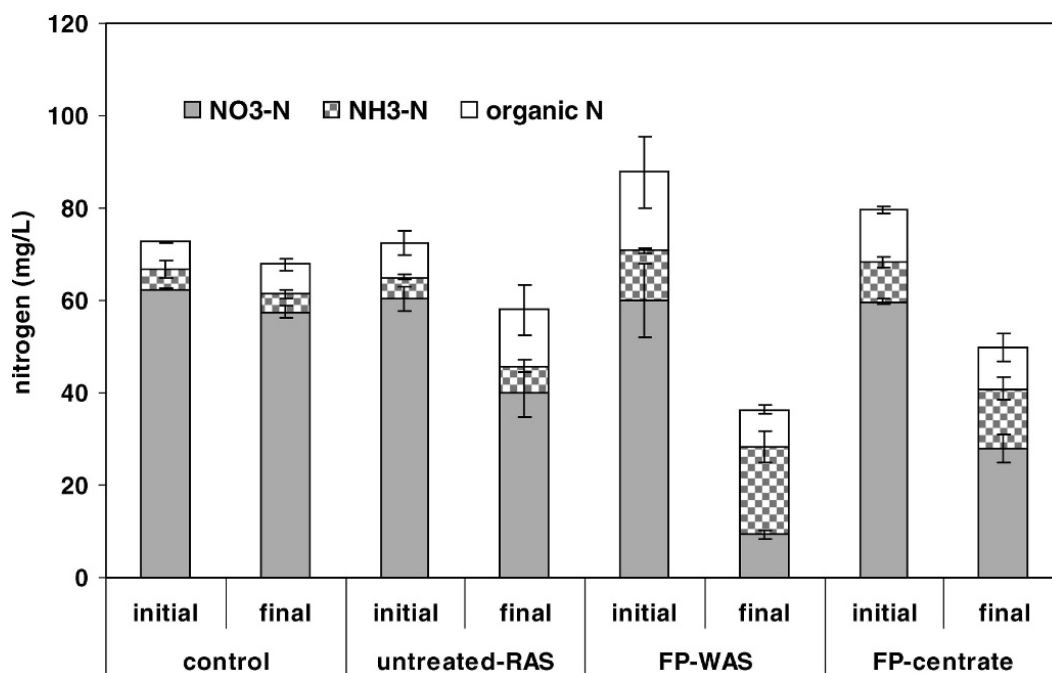


Figure 3—Changes in NO_3^- -N, semi-soluble NH_3 -N, and semi-soluble organic nitrogen during denitrification tests. NO_2^- -N was never detected; TKN is the sum of NH_3 -N and organic nitrogen; and total nitrogen is the sum of the three components. The conditions were the same for the two batch tests, and the data presented are the average (symbol) with range (bars).

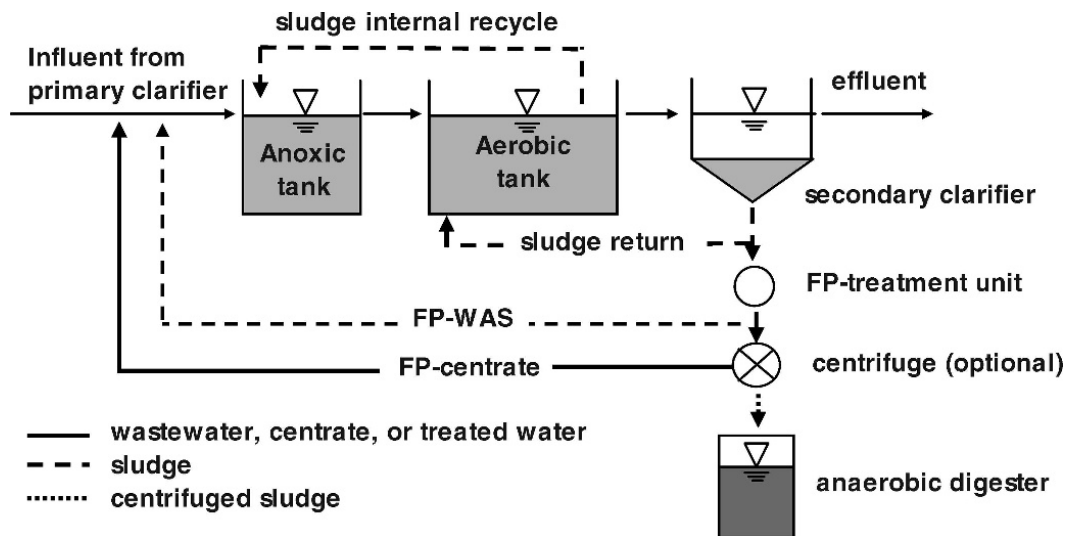


Figure 4—Schematic of how FP-WAS and FP-centrate would be used as an internal electron donor to drive pre-denitrification in a WWTP.

N. The final ratios of $\text{NH}_3\text{-N}$ -to-TN of FP-WAS and FP-centrate were 52% and 26%, respectively.

The results indicate that $\text{NH}_3\text{-N}$ and total Kjeldahl nitrogen (TKN) increased with the bioavailable organic matter when FP-WAS or FP-centrate was used as the electron donor for denitrification in the batch experiments. In addition, FP-WAS adds suspended solids. We evaluated the applicability of using FP-WAS or FP-centrate as an internal electron donor in WWTP using the pre-denitrification scenario shown in Figure 4. The FP-WAS can be used as an electron donor for denitrification (with or without being centrifuged) or sent to the anaerobic digester for methane production. Thus, using FP-WAS competes with methane production; the analysis in the Introduction indicates that the economic value of replacing a purchased electron donor is greater than that from the loss of methane. The FP-WAS returns TKN and suspended solids to the treatment train, while FP-centrate returns only TKN.

Table 5 shows the concentrations of TKN and TSS that would be added when FP-WAS or FP-centrate is added in a quantity large enough to denitrify an increment of 10 mg N/L of nitrate. The effects are proportional if the concentration of nitrate to be denitrified is smaller or larger. The added nitrogen load from using FP-WAS or FP-centrate would be approximately 1.1 or 1.6 mg N/L, respectively, and this corresponds to 11% ($1.1/10 \times 100\%$) or 16% ($1.6/10 \times 100\%$) of the NO_3^- -N removed. For comparison, the contribution of the nitrogen load that digester

centrate adds to a nitrifying activated sludge system is typically approximately 15%, even though it constitutes only 1% of the hydraulic load (van Dongen et al., 2001).

For TSS, the addition of FP-WAS adds approximately 357 mg/L to remove 10 mg-N/L. On the other hand, using FP-centrate adds no TSS, although the denitrification rate should be lower than when adding FP-WAS. Based on the increased ratio of phosphorus-to-SSCOD by FP treatment, which was 19 mg-TP/390 mg-SSCOD (Table 2), we also estimate increases of 0.7 and 1.1 mg/L total phosphorus for the use of FP-WAS and FP-centrate, respectively, as an electron donor.

Conclusions

We evaluated complementary facets of using WAS that has undergone FP pre-treatment as an internal electron donor to drive denitrification. The key findings are as follows:

- (1) SSCOD increased by 26 to 31 times after FP treatment compared with the untreated WAS. This was accompanied by parallel, but much smaller, increases of semi-soluble protein, sugar, TN, and TP.
- (2) The batch-test denitrification rate with FP-WAS ($0.25 \text{ g NO}_3^- \text{-N/g VSS}\cdot\text{d}$) was greater than for untreated-WAS ($0.05 \text{ g NO}_3^- \text{-N/g VSS}\cdot\text{d}$) and methanol ($0.15 \text{ g NO}_3^- \text{-N/g VSS}\cdot\text{d}$), while the denitrification rate of FP-centrate ($0.14 \text{ g NO}_3^- \text{-N/g VSS}\cdot\text{d}$) was comparable with methanol.

Table 5—Added nitrogen load and TSS concentration from using FP-WAS and FP-centrate as an electron donor.

Electron donor	$\Delta\text{NO}_3^- \text{-N}^{\text{a}}$	SSCOD/N ^b	Required SSCOD	N/SSCOD in electron donor ^c	TSS/SSCOD in electron donor ^d	Increased concentration	
						Nitrogen	TSS
FP-WAS	10 mg/L	1.5	15 mg/L	25/336	8000/336	1.1 mg/L	357 mg/L
FP-centrate	10 mg/L	2.3	23 mg/L	27/397	0/397	1.6 mg/L	0

^a This is to remove a unit of 10 mg-N/L by denitrification. Increases are proportional to this increment when NO_3^- -N removal differs from 10 mg-N/L.

^b This study (Table 3) = mg-COD/mg-N.

^c This study (Table 2) = mg-N/mg-COD.

^d This study (Table 2) = mg-TSS/mg-COD.

- (3) At the end of the batch tests, FP-WAS achieved the lowest TN and NO_3^- -N concentrations, but NH_4^+ -N was the highest, as a result of the release of organic nitrogen from the FP-treated biomass.
- (4) FP-centrate, which added no suspended solids as part the electron donor, had less release of NH_3 -N than FP-WAS at the end of denitrification tests, but it did not remove as much NO_3^- -N.
- (5) Although using FP-WAS or FP-centrate as an electron donor for denitrification recycles TKN that must be nitrified, the increase in nitrogen loading is only approximately 11 to 16% of the NO_3^- -N that is denitrified. It also returns approximately 0.7 to 1.1 mg P/L for the same conditions.
- (6) Although denitrification kinetics was slower when adding FP-centrate, compared with FP-WAS, it avoided recycling any suspended solids.

In summary, we provide proof of concept that FP-WAS has enough SSCOD with fast enough kinetics to drive denitrification when used as an added electron donor. The return of semi-soluble TN and TP is small compared with SSCOD, but the effects of added nitrogen and phosphorus loadings to the activated sludge process need to be considered.

Credits

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