

# N<sub>2</sub>O Emissions from Activated Sludge Processes, 2008–2009: Results of a National Monitoring Survey in the United States

JOON HO AHN,<sup>†</sup> SUNGPYO KIM,<sup>†,§</sup>  
HONGKEUN PARK,<sup>†</sup> BRIAN RAHM,<sup>†</sup>  
KRISHNA PAGILLA,<sup>‡</sup> AND  
KARTIK CHANDRAN<sup>\*,†</sup>

Department of Earth and Environmental Engineering,  
Columbia University, New York, New York 10027, and  
Department of Civil, Architectural and Environmental  
Engineering, Illinois Institute of Technology, Chicago, IL

Received December 18, 2009. Revised manuscript received  
April 20, 2010. Accepted May 4, 2010.

Despite recognition of the possible role of biological nitrogen removal (BNR) processes in nitrous oxide (N<sub>2</sub>O) emission, a measured database of N<sub>2</sub>O emissions from these processes at the national scale does not currently exist. This study focused on the quantification of N<sub>2</sub>O emissions at 12 wastewater treatment plants (WWTPs) across the United States using a newly developed U.S. Environmental Protection Agency (USEPA) reviewed protocol. A high degree of variability in field-scale measurements of N<sub>2</sub>O was observed, both across the WWTPs sampled and within each WWTP. Additionally, aerobic zones, which have hitherto not been considered in the USEPA approach of estimating N<sub>2</sub>O emissions, generally contributed more to N<sub>2</sub>O fluxes than anoxic zones from BNR reactors. These results severely qualify the conventional use of a single emission factor to “estimate” N<sub>2</sub>O emissions from BNR processes, solely by virtue of denitrification. Upon subjecting the nationwide data set to multivariate regression data mining, high nitrite, ammonium, and dissolved oxygen concentrations were positively correlated with N<sub>2</sub>O emissions from aerobic zones of activated sludge reactors. On the other hand, high nitrite and dissolved oxygen concentrations were positively correlated with N<sub>2</sub>O emissions from anoxic zones. Based on these results, it can be argued that activated sludge processes that minimize transient or permanent build up of ammonium or nitrite, especially in the presence of dissolved oxygen, are expected to have low N<sub>2</sub>O emissions.

## Introduction

Increasingly tighter regulations on total nitrogen removal from wastewater streams have spurred a global effort to develop and implement several cost-effective engineered biological nitrogen removal (BNR) strategies at point sources of nitrogen pollution such as wastewater treatment plants. Based on recent field-scale measurements, selected engi-

neered BNR facilities, while effective to varying degrees in reducing *aqueous* nitrogen pollution could emit up to 7% of the influent nitrogen load as *gaseous* nitrous oxide (N<sub>2</sub>O) and nitric oxide (NO), as described for the nitrifying B-stage of the Dokhaven-Sluisjesdijk WWTP in The Netherlands (1). Such emissions are deleterious to the environment. The greenhouse equivalence of N<sub>2</sub>O is about three hundred times that of carbon dioxide and both N<sub>2</sub>O and NO contribute to depletion of the ozone layer (2).

Although, from a fundamental perspective, N<sub>2</sub>O and NO are known intermediates in denitrification (3, 4) and nitrification (5–9), the net contribution of BNR processes to N<sub>2</sub>O emissions from wastewater treatment has only recently been acknowledged (10). Based on the latest U.S. Environmental Protection Agency (USEPA) report on sources and sinks of N<sub>2</sub>O from wastewater treatment operations, denitrification in anoxic zones is implicated as the dominant source of N<sub>2</sub>O from BNR activated sludge reactors (10). However, nitrification could also play a role in N<sub>2</sub>O generation and emission from WWTPs, especially under cycling between anoxic and aerobic conditions (1), as is common in BNR reactors. Therefore, N<sub>2</sub>O fluxes from aerobic zones of WWTPs might also need to be included in N<sub>2</sub>O emissions inventories.

At this point, while there is considerable debate and interest on the “potential” of constituent BNR processes for N<sub>2</sub>O generation and emission, there are few reports that systematically quantify such emissions from full-scale BNR operations (1, 11–15). Of these, only one has been conducted in the United States and focuses on a single non-BNR WWTP in New Hampshire (11). The remaining full-scale studies have been mostly conducted in Europe and have employed different methods for measuring N<sub>2</sub>O emissions including the use of grab samples (1, 11, 13–15) or online measurements (1, 12). Given the broad diversity of BNR and non-BNR configurations that exist in the United States, a more detailed N<sub>2</sub>O emissions database of WWTPs is needed, specifically obtained using a consistent protocol.

The principal motivation of conducting such a detailed monitoring campaign is the limitation in the currently used method to “estimate” the N<sub>2</sub>O emissions of WWTPs. The current method is based on emission factor values of 3.2 g N<sub>2</sub>O/population equivalents (PE)/year from non-BNR operations and 7.0 g N<sub>2</sub>O/PE/year for BNR operations (10, 11). Based on these two emission factors and an additional factor (0.5% of wastewater effluent nitrogen emitted as N<sub>2</sub>O), domestic wastewater treatment plants contribute approximately 3% to the total CO<sub>2</sub> footprint of waste management operations and 1.6% to total worldwide N<sub>2</sub>O emissions (10). However, both emission factors are based on a limited data set and may not be broadly representative.

Based on these outstanding gaps of knowledge, the specific objectives of this study were to 1. Develop a database of N<sub>2</sub>O emissions from different activated sludge process configurations using a standard protocol, and 2. Identify key factors that are correlated with N<sub>2</sub>O emissions from activated sludge.

## Materials and Methods

**Development and Application of a Protocol to Measure N<sub>2</sub>O Fluxes From BNR Processes.** The overall measurement campaign of this study was conducted using a recently developed USEPA reviewed protocol and quality assurance project plan for measuring liquid and gas phase N<sub>2</sub>O concentrations and their fluxes from activated sludge tanks (16). Commercially available replicas of the USEPA surface emission isolation flux chamber (SEIFC) with a circular cross-sectional area of 0.13 m<sup>2</sup> were used to measure gaseous N

\* Corresponding author phone: (212) 854 9027; fax: (212) 854 7081; e-mail: kc2288@columbia.edu.

<sup>†</sup> Columbia University.

<sup>‡</sup> Illinois Institute of Technology.

<sup>§</sup> Currently at: Department of Environmental Engineering, Korea University Sejong Campus, Korea.

**TABLE I. Summary of N<sub>2</sub>O Fluxes and Emission Factors Measured at Full-Scale WWTPs**

plant configuration	temp (°C)	reactor influent TKN load (g-N/day)	reactor effluent TN load (g-N/day)	Q (MGD)	% influent TKN emitted as N <sub>2</sub> O	% TN removed emitted as N <sub>2</sub> O	emission factor (g N <sub>2</sub> O/PE/yr)
separate-stage BNR	15 ± 0.48	1.8 × 10 <sup>6</sup>	3.6 × 10 <sup>5</sup>	23	0.03 ± 0.00	0.03 ± 0.01	1.2 ± 0.18
	23 ± 0.28	2.3 × 10 <sup>6</sup>	4.3 × 10 <sup>5</sup>	27	0.01 ± 0.00	0.01 ± 0.00	0.28 ± 0.13
four-stage Bardenpho	14 ± 0.26	8.6 × 10 <sup>5</sup>	1.7 × 10 <sup>5</sup>	7.8	0.16 ± 0.10	0.19 ± 0.12	9.8 ± 6.1
	23 ± 0.20	7.4 × 10 <sup>5</sup>	7.6 × 10 <sup>4</sup>	8.1	0.60 ± 0.29	0.66 ± 0.32	33 ± 16
step-feed BNR 1	19 ± 0.22	3.1 × 10 <sup>6</sup>	1.4 × 10 <sup>6</sup>	29	1.6 ± 0.83	2.9 ± 1.5	92 ± 47
	25 ± 0.28	2.9 × 10 <sup>6</sup>	9.4 × 10 <sup>5</sup>	30	0.62 ± 0.27	0.90 ± 0.39	33 ± 14
step-feed non-BNR	17 ± 0.12	8.6 × 10 <sup>6</sup>	4.4 × 10 <sup>6</sup>	71	0.18 ± 0.18	0.37 ± 0.36	13 ± 13
	26 ± 0.81	8.9 × 10 <sup>6</sup>	4.2 × 10 <sup>6</sup>	93	1.8 ± 0.79	3.3 ± 1.5	97 ± 43
separate centrate <sup>a</sup>	30 ± 2.3	8.8 × 10 <sup>6</sup>	5.5 × 10 <sup>6</sup>	2.0	0.24 ± 0.02	0.63 ± 0.06	<sup>a</sup>
	34 ± 0.32	8.5 × 10 <sup>6</sup>	4.2 × 10 <sup>6</sup>	1.6	0.54 ± 0.16	0.96 ± 0.32	<sup>a</sup>
plug-flow 1	11 ± 0.20	1.8 × 10 <sup>6</sup>	1.0 × 10 <sup>6</sup>	18	0.40 ± 0.14	0.92 ± 0.32	23 ± 7.9
	23 ± 0.46	1.8 × 10 <sup>6</sup>	7.3 × 10 <sup>5</sup>	15	0.41 ± 0.14	0.70 ± 0.24	28 ± 9.6
plug-flow 2	11 ± 0.41	6.3 × 10 <sup>5</sup>	4.0 × 10 <sup>5</sup>	8.7	0.62 ± 0.15	1.7 ± 0.41	26 ± 6.4
	22 ± 0.58	6.6 × 10 <sup>5</sup>	4.0 × 10 <sup>5</sup>	6.6	0.09 ± 0.03	0.22 ± 0.06	5.0 ± 1.4
MLE 1	26 ± 1.8	6.8 × 10 <sup>5</sup>	1.9 × 10 <sup>5</sup>	4.0	0.07 ± 0.04	0.09 ± 0.05	6.8 ± 3.5
MLE 2	26 ± 0.17	6.9 × 10 <sup>5</sup>	1.5 × 10 <sup>5</sup>	4.1	0.06 ± 0.02	0.07 ± 0.03	5.4 ± 2.0
step-feed BNR 2	29 ± 0.18	2.2 × 10 <sup>6</sup>	2.9 × 10 <sup>5</sup>	14	1.5 ± 0.02	1.7 ± 0.02	140 ± 1.2
oxidation ditch	19 ± 0.58	3.9 × 10 <sup>5</sup>	4.3 × 10 <sup>4</sup>	3.4	0.03 ± 0.01	0.03 ± 0.01	1.8 ± 0.77
step-feed BNR 3	24 ± 0.78	7.8 × 10 <sup>6</sup>	8.6 × 10 <sup>5</sup>	57	0.05 ± 0.03	0.06 ± 0.03	4.1 ± 2.2

<sup>a</sup> Flow normalized emission factors for centrate are inappropriate since centrate constitutes a very low flow rate (~ 1%), while containing up to 30% of the influent TKN load (19).

fluxes from activated sludge reactors. The SEIFC consists of a floating enclosed space through which exhaust gas can be collected in a real-time fashion. The SEIFC was employed to obtain headspace N<sub>2</sub>O concentrations and fluxes at different locations (aerated and nonaerated) in a treatment process. At each of these locations, online measurements of gaseous N<sub>2</sub>O concentrations were conducted over a 24 h period to address diurnal variability in gas emissions. Gaseous N<sub>2</sub>O concentrations were measured via infrared gas filter correlation (Teledyne API, San Diego, CA). Concomitantly, during each campaign, wastewater nitrogen species concentrations including influent, bioreactor and effluent TKN, ammonium (NH<sub>4</sub><sup>+</sup>-N), nitrite (NO<sub>2</sub><sup>-</sup>-N) and nitrate (NO<sub>3</sub><sup>-</sup>-N) were measured in the influent, effluent, and different locations directly next to the floating SEIFC at least six-times per day according to Standard Methods (17). Activated sludge mixed-liquor temperature, pH, dissolved oxygen (DO), soluble chemical oxygen demand (sCOD), mixed-liquor suspended solids (MLSS) and volatile suspended (VSS) concentrations were also measured using Standard Methods (17) directly next to the SEIFC location at the same time and frequency.

Advective flow rate of headspace gas through the SEIFC (Q<sub>emission</sub>, m<sup>3</sup>/min) in aerated and anoxic zones was measured approximately five times a day using a modification of ASTM method D1946. Briefly, a tracer gas consisting of 100 000 ppmv of helium (He) was introduced into the SEIFC at a known flow rate. He concentrations in the off-gas from the SEIFC were measured using a field gas-chromatograph equipped with thermal conductivity detection (GC-TCD). Q<sub>emission</sub> was computed based on dilution of the He tracer in the off-gas relative to the influent (16).

The net mass flux of N<sub>2</sub>O (J<sub>N<sub>2</sub>O,SEIFC</sub>, mg/min-m<sup>2</sup>) from the flux chamber equaled (Q<sub>emission</sub>C/A<sub>SEIFC</sub>), where C is the gas concentration (mg/m<sup>3</sup>) and A<sub>SEIFC</sub> is the cross-sectional area of the SEIFC (m<sup>2</sup>). The overall mass emission (g N<sub>2</sub>O-N/d) was calculated by multiplying the mass flux (J<sub>N<sub>2</sub>O,SEIFC</sub>) and the surface area of the activated sludge bioreactor (A<sub>AS</sub>). The proportion of N<sub>2</sub>O emitted from each WWTP was computed based on the measured mass flux from each zone in the WWTP normalized to the daily influent total Kjeldahl nitrogen (TKN) loading (mass/mass) or daily TKN load processed (which refers to the difference between the influent TKN load and effluent ammonia load, mass/mass).

**Calculation of N<sub>2</sub>O Emission Factors.** N<sub>2</sub>O emission factors were computed by normalizing the total reactor N<sub>2</sub>O mass flux to the unit population equivalent flow rate (100 gal/PE/day) and were expressed in units consistent with the USEPA inventory report (g N<sub>2</sub>O/PE/year) (10). For aerobic zones, the helium-based advective gas-flow data were correlated to plant-recorded airflow rates for any given zone via linear regression and used to calculate diurnal N<sub>2</sub>O emission factors. For anoxic (nonaerated) zones lacking associated plant airflow data, the average of the experimentally obtained helium-based gas flow rates was used to calculate diurnal N<sub>2</sub>O emission factors.

**Operating Characteristics of Wastewater Treatment Processes Sampled.** The 12 WWTPs at which N<sub>2</sub>O emissions were measured employed a variety of BNR and non-BNR process configurations (Table I). Although the specific identities of the WWTPs cannot be described, they were broadly distributed around the Northeast (4), Mid-Atlantic (2), Midwest (2) and Southwest (4) regions of the United States. Additionally, 7 out of the 12 processes were sampled at minimum and maximum annual wastewater temperatures to examine seasonal temperature impacts on N<sub>2</sub>O generation and emission.

**Correlation Between WWTP Operating Conditions and N<sub>2</sub>O Emissions.** Possible links between WWTP operating conditions and N<sub>2</sub>O emission fluxes were examined via multivariate linear regression modeling using SAS (Cary, NC). For aerobic zones, N<sub>2</sub>O fluxes were correlated with sampling location-specific pH, activated sludge mixed-liquor temperature, DO, NH<sub>4</sub><sup>+</sup>-N, and NO<sub>2</sub><sup>-</sup>-N concentrations and interactive combinations thereof. For anoxic zones, N<sub>2</sub>O fluxes were correlated with sampling location-specific sCOD, pH, activated sludge mixed-liquor temperature, DO, NO<sub>2</sub><sup>-</sup>-N, and NO<sub>3</sub><sup>-</sup>-N concentrations and interactive combinations thereof. Assumptions of state variable normality and equal variance were evaluated using error residual and covariance plots (not shown). The variables not normally distributed were log-transformed, verified for normality and equal-variance and subsequently used for regression modeling. Time points where all state variables had not been measured simultaneously were not included in regression analysis.

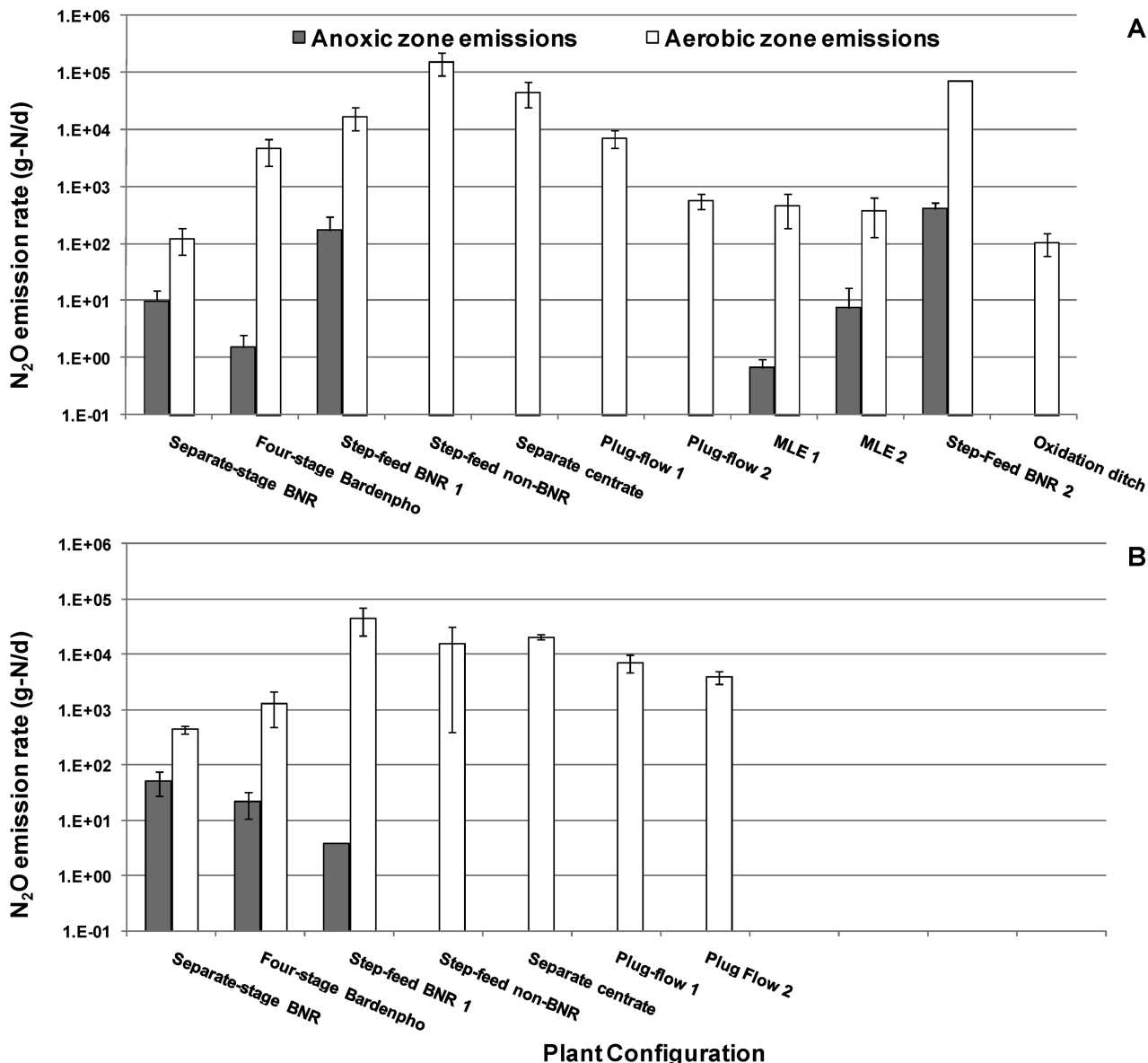


FIGURE 1.  $N_2O$  emissions from aerobic and anoxic zones in different WWTPs measured at high (A) and low (B) temperatures. Specific temperatures described in Table I. Step-feed BNR 3 is not included since the emissions from the covered aerobic and anoxic zones could not be distinctly measured.

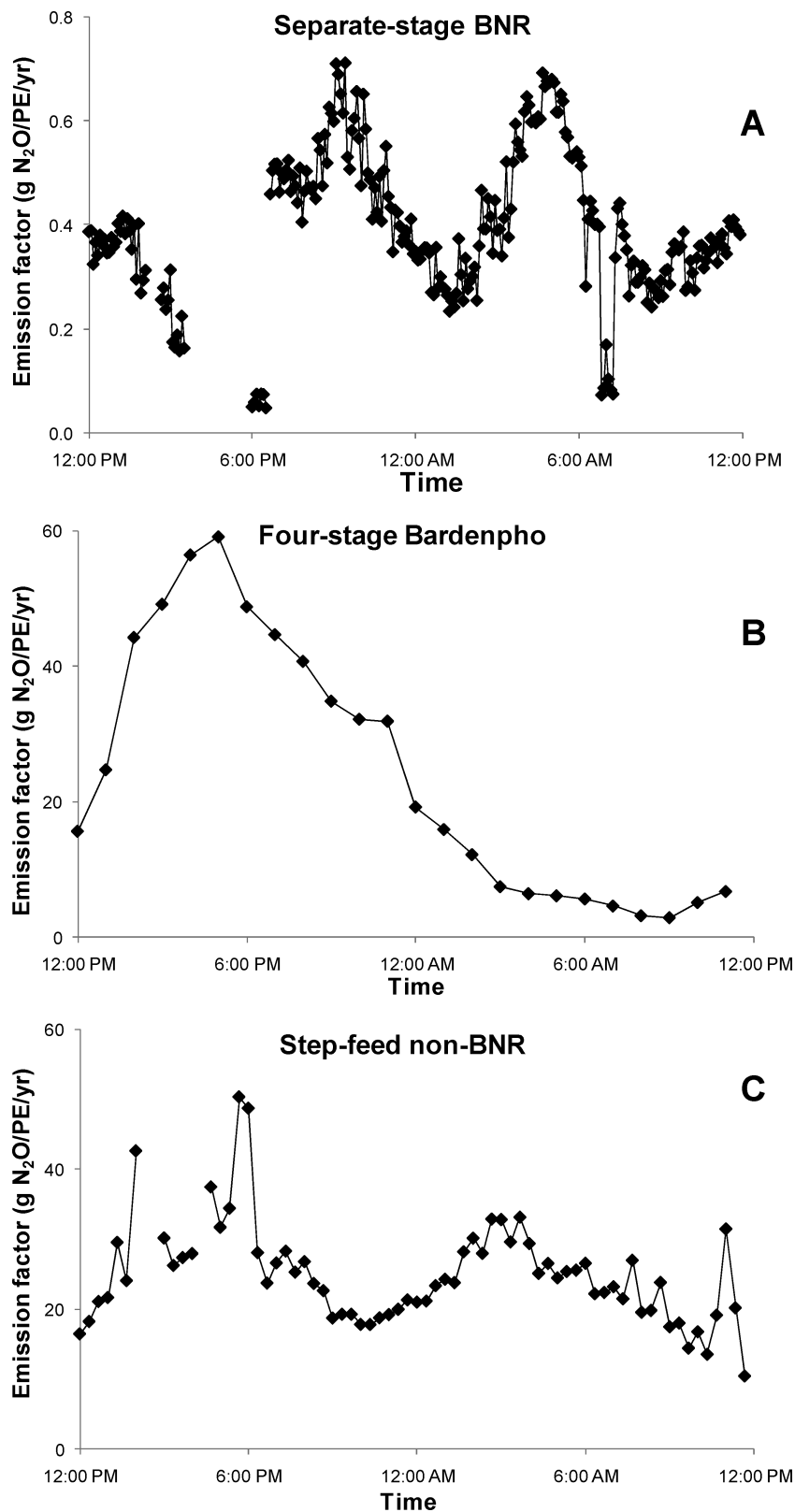
## Results and Discussion

**$N_2O$  Emission Fluxes from Activated Sludge Processes.** A wide range of  $N_2O$  emissions was measured across the 12 WWTPs operated at different temperatures, configurations and influent TKN loads (Table I). On average,  $N_2O$  emission fractions varied from 0.01 to 1.8% or 0.01 to 3.3%, when normalized to influent TKN load or influent TKN load processed, respectively. These emission fractions were on the lower end of the range reported by previous studies, which varied between 0 and 15% of influent TKN load (1, 11–15).

In general,  $N_2O$  emissions in aerated zones were higher than those in nonaerated zones (Figure 1). Therefore, the currently held premise that  $N_2O$  emissions from WWTPs mostly occur in the anoxic zones (10) is not accurate. Higher emissions from aerobic zones can be attributed to three possible phenomena: increased air-stripping of  $N_2O$ ,  $N_2O$  production due to oxygen inhibited denitrification or  $N_2O$  production by nitrification during changes in aeration. While it was not possible to infer the specific contribution of these phenomena to overall emissions from our data set, attempts were indeed made to identify links between principal

operating parameters and measured emissions (described in the modeling section). Several WWTPs did not have dedicated anoxic zones (Figure 1), although transient DO limiting or anoxic conditions were indeed observed in them, for instance in the plug-flow or oxidation-ditch processes (shown in the Supporting Information (SI)).

Computed flow-normalized emission factors also varied in a wide range (Table I), and for several processes, were statistically higher ( $\alpha = 0.05$ ) than currently used values of 3.2 g  $N_2O$ /PE/yr (non-BNR processes (11)) or 7.0 g  $N_2O$ /PE/yr (BNR processes (10)). Emission factors from the separate-stage BNR and oxidation ditch process were statistically lower, ( $\alpha = 0.05$ ) and those from the two MLE processes were statistically not dissimilar, ( $\alpha = 0.05$ ) relative to the current estimates (10). The centrate treatment process received the centrifuge supernatant of the anaerobic digestion recycle stream and was operated to partially convert the influent  $NH_4^+$ -N load to  $NO_2^-$ -N. Such operation is intended to reduce oxygen requirements for nitrification and organic carbon requirements for subsequent denitrification (as a variant of the single reactor high-activity ammonia removal over nitrite, SHARON process (18)). Emission factors for the



**FIGURE 2.** Time-varying  $N_2O$  emission factors for three select processes sampled in this study at  $23 \pm 0.28$  °C (A),  $23 \pm 0.20$  °C (B), and  $26 \pm 0.81$  °C (C).

separate centrate treatment process were not computed because centrate streams have disproportionately low flow rates compared to their TKN concentrations (19), which lead to incomparably high emission factor values. A high degree of diurnal variability in emission factors from the overall processes sampled was also observed (shown for three select processes in Figure 2) and could be linked to diurnal variations

in influent N-loading (data not shown) as reported previously (20). Based on the observed variability either diurnally or across the range of WWTPs sampled, the use of a “single” universal emission factor to calculate  $N_2O$  emissions from all wastewater treatment processes is inadequate and strongly discouraged. Further, due to significant differences in local or global wastewater composition (19), the use of flow

**TABLE II. Factors Correlating With N<sub>2</sub>O Emission Fluxes (g N<sub>2</sub>O—N/day) from Aerobic Zones**

variable	parameter estimate	standard error	t value	Pr >  t
intercept	6.1	0.48	13	0.00
log (ammonium)	1.0	0.25	4.1	0.00
log (nitrite)	0.60	0.16	3.7	0.00
log (DO)	0.59	0.18	3.2	0.00
log (ammonium)-log(nitrite)	0.18	0.08	2.3	0.03

normalized emission factors to describe N<sub>2</sub>O emissions is also not recommended.

Given that generation and emission of N<sub>2</sub>O and NO from biological wastewater treatment processes is ultimately driven by different microbial pathways and reactions, a better approach would be to understand the triggers and factors that promote such emissions and develop strategies toward their minimization. The results of a data mining approach to correlate principal factors with N<sub>2</sub>O emissions from the nationwide data set are described next.

**Model Based Discrimination of Process Operating Conditions Contributing to N<sub>2</sub>O Emission.**

*Aerobic zones.* Based on multivariate regression modeling, the factors positively correlated with N<sub>2</sub>O emissions from aerobic zones, were NH<sub>4</sub><sup>+</sup>-N, NO<sub>2</sub><sup>-</sup>-N, and DO concentrations (isolated effect), and NH<sub>4</sub><sup>+</sup>-N and NO<sub>2</sub><sup>-</sup>-N concentrations (interactive effect, Table II). From a fundamental perspective, nitrite is one of the most important factors contributing to N<sub>2</sub>O production from nitrification for different ammonia oxidizing bacteria including *Nitrosomonas europaea* (21–23) and several strains of *Nitrosospira* spp. (24), which are expected to be more abundant in activated sludge (25). Thus, correlation of NO<sub>2</sub><sup>-</sup>-N concentrations with aerobic zone N<sub>2</sub>O emissions is consistent with basic biochemical principles. Although it was previously proposed that low DO concentrations were directly responsible for nitrification-related N<sub>2</sub>O emissions (26), recent results suggest that N<sub>2</sub>O is generated by *recovery* from low DO conditions rather than *imposition* thereof (27). In fact, abrupt increases in DO concentrations in the presence of NH<sub>4</sub><sup>+</sup> can also lead to transient accumulation of NO<sub>2</sub><sup>-</sup>, which can in turn result in NO and N<sub>2</sub>O generation from nitrification (27, 28). The magnitude of nitrification driven N<sub>2</sub>O emissions also depends directly on NH<sub>4</sub><sup>+</sup>-N concentrations (27). Therefore, the positive correlation of NH<sub>4</sub><sup>+</sup>-N and DO concentrations individually with N<sub>2</sub>O emissions in the aerobic zones is in congruence with this new understanding of aerobic N<sub>2</sub>O production from nitrification (27). The positive interactive correlation between NH<sub>4</sub><sup>+</sup> and NO<sub>2</sub><sup>-</sup> concentrations and N<sub>2</sub>O emissions suggests high N<sub>2</sub>O emissions from aerobic zone locations, with *simultaneously* high concentrations of both NH<sub>4</sub><sup>+</sup> and NO<sub>2</sub><sup>-</sup>. This interactive correlation also points to N<sub>2</sub>O generation mechanisms from nitrification, wherein NH<sub>4</sub><sup>+</sup> and NO<sub>2</sub><sup>-</sup> can act as cosubstrates, (as described for *Nitrosomonas europaea* (21–23) and *Nitrosospira* spp. (24)).

It should be noted that while nitrification dominates N-cycling in aerobic zones, the possibility of denitrification within activated sludge flocs even under “bulk” aerobic conditions cannot be discounted (29). Interestingly, as with nitrification, denitrification driven N<sub>2</sub>O emissions are also caused by high DO and NO<sub>2</sub><sup>-</sup>-N concentrations (4, 30–33), among other factors. Thus, aerobic zone N<sub>2</sub>O emissions might be linked to both nitrification and denitrification, although it is not possible to differentiate between the two using bulk headspace measurements alone.

*Anoxic Zones.* Several factors have been implicated in N<sub>2</sub>O emissions from denitrifying bioreactors, such as low pH (34),

**TABLE III. Factors Correlating with N<sub>2</sub>O Emission Fluxes (g N<sub>2</sub>O—N/day) from Anoxic Zones**

variable	parameter estimate	standard error	t value	Pr >  t
intercept	-1.2	0.89	-1.3	0.21
log(DO)-log(nitrite)	0.67	0.25	2.7	0.01

short solids retention time (35), organic carbon limitation (35–37), DO inhibition (31, 33), and NO<sub>2</sub><sup>-</sup>-N inhibition (4, 30, 32). Inhibition of denitrification by high DO concentrations could also lead to NO<sub>2</sub><sup>-</sup>-N build-up, indirectly leading to N<sub>2</sub>O emissions (35, 38). Thus, the positive correlation of DO and NO<sub>2</sub><sup>-</sup>-N concentrations with N<sub>2</sub>O emissions (Table III) is consistent with known mechanisms of denitrification-related N<sub>2</sub>O production. Soluble COD concentrations in anoxic zones were mostly nonlimiting (29) (provided in the SI) and expectedly did not correlate with N<sub>2</sub>O emissions.

The regression model did not correlate temperature and N<sub>2</sub>O fluxes in either aerobic or anoxic zones (Tables II–III). A possible reason could be that N<sub>2</sub>O fluxes are indirectly governed by temperature through manifestation in NH<sub>4</sub><sup>+</sup>-N, NO<sub>2</sub><sup>-</sup>-N, or DO concentrations, as described above. For instance, in a process not designed or under-designed for nitrification, high NH<sub>4</sub><sup>+</sup>-N concentrations are more distributed over the entire bioreactor volume during low temperatures (due to lower nitrification kinetics and lower nitrifying bacteria concentrations). Additionally, the lower temperatures also could potentially increase the solubility of oxygen in the mixed-liquor (39). Given that nonlimiting NH<sub>4</sub><sup>+</sup>-N and DO concentrations lead to N<sub>2</sub>O (27) (and results from correlation analysis in Table II), N<sub>2</sub>O emissions are expected to be higher in such systems at low temperature. In contrast, for systems that are designed to completely nitrify throughout the year (and have limiting reactor and effluent NH<sub>4</sub><sup>+</sup>-N concentrations), the emissions of N<sub>2</sub>O are expected to be higher during higher temperature (due to higher overall kinetics of both oxidative and reductive nitrogen transformations). Notwithstanding the above argument, the actual configuration of the bioreactor also contributes to N<sub>2</sub>O emissions by virtue of localized concentrations and rate-determining regimes. Thus, it might be more illustrative to correlate N<sub>2</sub>O emissions with localized N-species and DO concentrations rather than with operating temperature in general.

While several factors relevant to activated sludge processes including NH<sub>4</sub><sup>+</sup>, NO<sub>2</sub><sup>-</sup>, DO, temperature, COD limitation can all lead to N<sub>2</sub>O emissions, it may be difficult to discern their specific contributions from highly noisy full-scale data from multiple plants, each operated under different modes and temperatures. Therefore, it is noteworthy that N<sub>2</sub>O emissions in aerobic and anoxic zones could be correlated with select specific factors and explained via mechanistic arguments. Further, all model parameter estimates except the intercept of the anoxic zone were statistically valid at the α = 0.05 level (Tables II–III). Nevertheless, it should be noted that the developed model is not a conclusive mechanistic descriptor of causative biological processes contributing to N<sub>2</sub>O emissions from activated sludge. Caution is also advised with respect to predicting emissions on the basis of process configuration alone. The factors correlating with N<sub>2</sub>O emissions, (NH<sub>4</sub><sup>+</sup>-N, NO<sub>2</sub><sup>-</sup>-N, and DO concentrations) are inherently linked with process parameters such as TKN loadings, solids residence time (SRT) and wastewater composition. Therefore, the propensity for N<sub>2</sub>O emissions of any given WWTP configuration can only be evaluated within the joint framework of its process operation and performance characteristics.

**N<sub>2</sub>O Emissions From WWTPs and Sustainable BNR Design and Operations.** The sustainable management of nitrogenous pollution is one of the grand challenges of the National Academy of Engineering (<http://www.engineeringchallenges.org/>). Thus, from an engineering perspective, it would be beneficial to develop sound strategies for BNR design and operation that minimize both *aqueous* and *gaseous* nitrogen emissions. Based on this study, the measured emission factors from some BNR processes (Table I) were at least 1 order of magnitude higher than current estimates (10). However, given that the emission factors from other processes, were clearly lower than current emission factor estimates (Table I), the prospect of engineering based minimization N<sub>2</sub>O emissions from WWTPs appears to be feasible.

It should nevertheless be strongly emphasized that the results of this study do not implicate biological wastewater treatment and biological nutrient removal as prime contributors to atmospheric N<sub>2</sub>O fluxes. Even if the emission factors of all WWTPs were to hypothetically be universally an order of magnitude higher (which is highly unlikely, as shown in this study), domestic wastewater treatment would still contribute just 14% of the overall N<sub>2</sub>O emissions (using the same figures for N<sub>2</sub>O emissions from other sources, from (10)). Clearly, compromising domestic wastewater treatment and nutrient removal for the sake of reducing N<sub>2</sub>O emissions could have far more negative implications to water quality and human health.

More than a greenhouse or ozone depletion perspective, low N<sub>2</sub>O emissions from BNR processes can also be an indication of stable nitrification and denitrification (27, 40). Based on regression modeling results (Tables II-III), BNR processes that avoid high NH<sub>4</sub><sup>+</sup>-N, NO<sub>2</sub><sup>-</sup>-N, and DO concentrations and transients thereof are expected to generate less N<sub>2</sub>O, from both nitrification and denitrification. High NH<sub>4</sub><sup>+</sup>-N, DO and NO<sub>2</sub><sup>-</sup>-N concentrations are typically encountered in under-designed and overaerated nitrification reactors (29). Therefore, aerobic processes that avoid incomplete or intermittent nitrification and overaeration are expected to have lower N<sub>2</sub>O emissions. Additionally, processes that rely on more uniform spatial DO profiles to promote simultaneous nitrification and denitrification such as the oxidation ditch sampled herein, are likely to have low N<sub>2</sub>O emissions. Such processes avoid repeated switching between aerobic and anoxic conditions, which in conjunction with high NH<sub>4</sub><sup>+</sup>-N and DO concentrations are a prime cause for nitrification driven N<sub>2</sub>O emissions (27). An additional strategy to minimize N<sub>2</sub>O emissions could be to minimize the peaking factor of influent nitrogen loading to activated sludge via flow equalization (20). While flow equalization of primary effluent may be difficult because of the high flow rates, it might be possible in select WWTPs to blend high nitrogen containing centrate with primary effluent to equalize influent nitrogen loads.

This study is the first of its kind in the United States and there are only two other similar studies being conducted, in The Netherlands (Prof. M.C.M van Loosdrecht, Delft University of Technology, personal communication) and Australia (Prof. Z. Yuan, University of Queensland, personal communication). To facilitate a global monitoring effort, the protocol developed in this study has been shared upon request with additional groups in Spain, Portugal, Germany, and Belgium, which are just beginning similar monitoring programs. It is expected that broad application of the protocol will allow WWTPs to quantify their N<sub>2</sub>O emissions using a standard approach and to ultimately engineer environmentally sustainable BNR operating strategies and configurations.

## Acknowledgments

This study is supported by the Water Environment Research Foundation. The monitoring campaigns will be continued over the next few years to broaden the sampling pool of WWTPs including those that will transition to BNR operations in the time being. As the monitoring progresses, updates to the emissions database will be presented on the website ([www.columbia.edu/~kc2288/CCR](http://www.columbia.edu/~kc2288/CCR)) and in follow up publications.

## Supporting Information Available

Process schematics of WWTP reactors and the process configuration table. This material is available free of charge via the Internet at <http://pubs.acs.org>.

## Literature Cited

- (1) Kampschreur, M. J.; van der Star, W. R. L.; Wielders, H. A.; Mulder, J. W.; Jetten, M. S. M.; van Loosdrecht, M. C. M. Dynamics of nitric oxide and nitrous oxide emission during full-scale reject water treatment. *Water Res.* **2008**, *42* (3), 812–826.
- (2) Ravishankara, A. R.; Daniel, J. S.; Portmann, R. W. Nitrous oxide (N<sub>2</sub>O): The dominant ozone-depleting substance emitted in the 21st century. *Science* **2009**, *326*, 123–125.
- (3) Knowles, R. Denitrification. *Microbiol. Rev.* **1982**, *46* (1), 43–70.
- (4) Zumft, W. G. Cell biology and molecular basis of denitrification. *Microbiol. Mol. Biol. Rev.* **1997**, *61* (4), 533–616.
- (5) Anderson, I. C.; Levine, J. S. Relative rates of nitric oxide and nitrous oxide production by nitrifiers, denitrifiers and nitrate respirers. *Appl. Environ. Microbiol.* **1986**, *51* (5), 938–945.
- (6) Anderson, I. C.; Poth, M.; Homstead, J.; Burdige, D. A comparison of NO and N<sub>2</sub>O production by the autotrophic nitrifier *Nitrosomonas europaea* and the heterotrophic nitrifier *Alcaligenes faecalis*. *Appl. Environ. Microbiol.* **1993**, *59* (11), 3525–3533.
- (7) Kester, R. A.; de Boer, W.; Laanbroek, H. J. Production of NO and N<sub>2</sub>O by pure cultures of nitrifying and denitrifying bacteria during changes in aeration. *Appl. Environ. Microbiol.* **1997**, *63*, 3872–3877.
- (8) Ritchie, G. A. F.; Nicholas, D. J. D. Identification of the sources of nitrous oxide produced by oxidative and reductive processes in *Nitrosomonas europaea*. *Biochem. J.* **1972**, *126*, 1181–1191.
- (9) Stuvan, R.; Vollmer, M.; Bock, E. The impact of organic matter on nitric oxide formation by *Nitrosomonas europaea*. *Arch. Microbiol.* **1992**, *158* (6), 439–443.
- (10) *Inventory of U.S. Greenhouse Gas Emissions and Sinks: 1990–2006*, EPA 430-R-08-005; U. S. Environmental Protection Agency: Washington, DC, 2009.
- (11) Czepiel, P.; Crill, P.; Harriss, R. Nitrous oxide emissions from municipal wastewater treatment. *Environ. Sci. Technol.* **1995**, *29* (9), 2352–2356.
- (12) Kimochi, Y.; Inamori, Y.; Mizuochi, M.; Xu, K.-Q.; Matsumura, M. Nitrogen removal and N<sub>2</sub>O emission in a full-scale domestic wastewater treatment plant with intermittent aeration. *J. Ferment. Bioeng.* **1998**, *86* (2), 202–206.
- (13) Sommer, J.; Ciplak, A.; Sumer, E.; Benckiser, G.; Ottow, J. C. G. Quantification of emitted and retained N<sub>2</sub>O in a municipal wastewater treatment plant with activated sludge and nitrification-denitrification units. *Agrobiol. Res.* **1998**, *51* (1), 59–73.
- (14) Sümer, E.; Weiske, A.; Benckiser, G.; Ottow, J. C. G. Influence of environmental conditions on the amount of N<sub>2</sub>O released from activated sludge in a domestic waste water treatment plant. *Cell. Mol. Life Sci.* **1995**, *51* (4), 419–422.
- (15) Wicht, H.; Beier, M. N<sub>2</sub>O emission aus nitrifizierenden und denitrifizierenden Klaranlagen. *Korresp. Abwasser* **1995**, *42* (3), 404–406, 411–413.
- (16) Chandran, K. *Characterization of Nitrogen Greenhouse Gas Emissions from Wastewater Treatment BNR operations: Field Protocol with Quality Assurance Plan*; Water Environment Research Foundation: Alexandria, VA, 2009.
- (17) Eaton, A. D.; Clesceri, L. S.; Greenberg, A. E., *Standard Methods for the Examination of Water and Wastewater*. ed.; APHA, AWWA and WEF: Washington DC, 2005.
- (18) Hellinga, C.; Schellen, A. A. J. C.; Mulder, J. W.; van Loosdrecht, M. C. M.; Heijnen, J. J. The Sharon process: An innovative method for nitrogen removal from ammonium-rich waste water. *Water Sci. Technol.* **1998**, *37* (9), 135–142.
- (19) Tchobanoglous, G.; Burton, F. L.; Stensel, H. D., *Metcalf and Eddy Wastewater Engineering: Treatment and Reuse*. 4th ed.; McGraw Hill: New York, NY, 2003.

- (20) Ahn, J.-H.; Kim, S.; Pagilla, K.; Katehis, D.; Chandran, K. In *Spatial and Temporal Variability in N<sub>2</sub>O Generation and Emission from Wastewater Treatment Plants*; Nutrient Removal Conference, Water Environment Federation: Washington D.C., 2009.
- (21) Beaumont, H. J. E.; Lens, S. I.; Westerhoff, H. V.; van Spanning, R. J. M. Novel *nirK* cluster genes in *Nitrosomonas europaea* are required for NirK-dependent tolerance to nitrite. *J. Bacteriol.* **2005**, *187* (19), 6849–6851.
- (22) Beaumont, H. J. E.; van Schooten, B.; Lens, S. I.; Westerhoff, H. V.; van Spanning, R. J. M. *Nitrosomonas europaea* expresses a nitric oxide reductase during nitrification. *J. Bacteriol.* **2004**, *186* (13), 4417–4421.
- (23) Beaumont, H. J. E.; Lens, S. I.; Reijnders, W. N. M.; Westerhoff, H. V.; van Spanning, R. J. M. Expression of nitrite reductase in *Nitrosomonas europaea* involves NsrR, a novel nitrite-sensitive transcription repressor. *Mol. Microbiol.* **2004**, *54* (1), 148–158.
- (24) Shaw, L. J.; Nicol, G. W.; Smith, Z.; Fear, J.; Prosser, J. I.; Baggs, E. M. *Nitrosospira* spp. can produce nitrous oxide via a nitrifier denitrification pathway. *Environ. Microbiol.* **2006**, *8* (2), 214–222.
- (25) Coskuner, G.; Curtis, T. P. *In-situ* characterization of nitrifiers in an activated sludge plant: detection of *Nitrobacter*spp. *J. Appl. Microbiol.* **2002**, *93* (3), 431–437.
- (26) Tallec, G.; Garnier, J.; Billen, G.; Gousailles, M. Nitrous oxide emissions from secondary activated sludge in nitrifying conditions of urban wastewater treatment plants: Effect of oxygenation level. *Water Res.* **2006**, *40* (15), 2972–2980.
- (27) Yu, R.; Kampschreur, M. J.; van Loosdrecht, M. C. M.; Chandran, K. Mechanisms and specific directionality of autotrophic nitrous oxide and nitric oxide generation during transient anoxia. *Environ. Sci. Technol.* **2010**, *44* (4), 1313–1319.
- (28) Kampschreur, M. J.; Tan, N. C. G.; Kleerebezem, R.; Picioreanu, C.; Jetten, M. S. M.; Loosdrecht, M. C. M. v. Effect of dynamic process conditions on nitrogen oxides emission from a nitrifying culture. *Environ. Sci. Technol.* **2008**, *42* (2), 429–435.
- (29) Grady, C. P. L. J.; Daigger, G. T.; Lim, H. C., *Biological Wastewater Treatment*, 2nd ed.; Marcel Dekker: New York, 1999.
- (30) Korner, H.; Zumft, W. G. Expression of denitrification enzymes in response to the dissolved oxygen level and respiratory substrate in continuous culture of *Pseudomonas stutzeri*. *Appl. Environ. Microbiol.* **1989**, *55*, 1670–1676.
- (31) Park, K. Y.; Inamori, Y.; Mizuochi, M.; Ahn, K. H. Emission and control of nitrous oxide from a biological wastewater treatment system with intermittent aeration. *Soc. Biotechnol., Jpn.* **2000**, *90* (3), 247–252.
- (32) Schulthess, R. v.; Kuhni, M.; Gujer, W. Release of nitric and nitrous oxides from denitrifying activated sludge. *Water Res.* **1995**, *29* (1), 215–226.
- (33) Tallec, G.; Garnier, J.; Billen, G.; Gousailles, M. Nitrous oxide emissions from denitrifying activated sludge of urban wastewater treatment plants, under anoxia and low oxygenation. *Bioresour. Technol.* **2008**, *99* (7), 2200–2209.
- (34) Focht, D. D. The effect of temperature, pH and aeration on the production of nitrous oxide and gaseous nitrogen: A zero-order kinetic model. *Soil Science* **1974**, *118* (8), 173–179.
- (35) Hanaki, K.; Hong, Z.; Matsuo, T. Production of nitrous oxide gas during denitrification of wastewater. *Water Sci. Technol.* **1992**, *26* (1–12), 1027–1036.
- (36) von Schultess, R.; Kuhni, M.; Gujer, W. Release of nitric and nitrous oxides from denitrifying activated sludge. *Water Res.* **1995**, *29* (1), 215–226.
- (37) von Schultess, R.; Wild, D.; Gujer, W. Nitric and nitrous oxides from denitrifying activated sludge at low oxygen concentration. *Water Sci. Technol.* **1994**, *30* (8), 123–132.
- (38) Zhou, Y.; Pijuan, M.; Zeng, R. J.; Yuan, Z. Free nitrous acid inhibition on nitrous oxide reduction by a denitrifying-enhanced biological phosphorus removal sludge. *Environ. Sci. Technol.* **2008**, *42* (22), 8260–8265.
- (39) Stumm, W.; Morgan, J. J., *Aquatic Chemistry Chemical Equilibria and Rates in Natural Waters*, 3rd ed.; John Wiley and Sons: New York, 1996.
- (40) Butler, M. D.; Wang, Y. Y.; Cartmell, E.; Stephenson, T. Nitrous oxide emissions for early warning of biological nitrification failure in activated sludge. *Water Res.* **2009**, *43* (5), 1265–1272.

ES903845Y