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Seasonal and diurnal variability of N_2O emissions from a full-scale municipal wastewater treatment plant

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HIGHLIGHTS

- Unique dataset of long-term nitrous oxide emission from activated sludge tanks
- Emission exhibited pronounced diurnal variability, superimposed on seasonal trend
- Seasonal nitrous oxide emission trend correlated with daily nitrite peaks
- Emission's diurnal trend suggests suboptimal oxygen concentrations as cause

GRAPHICAL ABSTRACT

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ABSTRACT

During nitrogen removal in conventional activated sludge processes, nitrous oxide can be emitted. With a global warming potential of 298 CO_2-equivalents it is an important greenhouse gas that affects the sustainability of wastewater treatment. The present study reports nitrous oxide emission data from a 16 month monitoring campaign on a full-scale municipal wastewater treatment. The emission demonstrated a pronounced diurnal and seasonal variability. This variability was compared with the variability of a number of process variables that are commonly available on a municipal wastewater treatment plant. On a seasonal timescale, the occurrence of peaks in the nitrite concentration correlated strongly with the emission. The diurnal trend of the emission coincided with the diurnal trend of the nitrite and nitrate concentrations in the tank, suggesting that suboptimal oxygen concentrations may induce the production of nitrous oxide during both nitrification and denitrification. This study documents an unprecedented dataset that could serve as a reference for further research.

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1. Introduction

Since about two decades it has been recognised that nitrous oxide (N_2O) is emitted during the biological treatment of wastewater (Hanaki et al., 1992). Considering its global warming potential of 298 CO_2-equivalents over a hundred year time horizon, the emission of nitrous oxide can contribute significantly to the climate footprint of a wastewater treatment plant (IPCC, 2013). At the WWTP that is also the location for the present study, nitrous oxide was found to make up 78.4% of the plants climate footprint, expressed as CO_2-equivalents, the remainder of the emissions being methane (13.5%) and the indirect carbon dioxide related to the plant's energy consumption (8.1%) (Daelman et al., 2013b). Besides a considerable greenhouse gas, nitrous oxide is currently also the most important ozone-depleting substance emitted (Ravishankara et al., 2009). In literature, emission values as high as 25.4% of the nitrogen load of the WWTP have been reported and from Table 1 it is obvious that there is a huge spread of the reported emissions. The question remains to what extent this variability is caused by differences in the real emission or by differences in sampling method (Daelman et al., 2013a).

Nitrous oxide is produced and emitted during biological nitrogen removal from wastewater through nitrification and denitrification. Schreiber et al. (2012) give an extensive overview of the biological pathways and the chemical reactions that can lead to the formation of nitrous oxide. Nitrous oxide is produced biologically as an intermediate metabolite by heterotrophic denitrifying bacteria and as a side product by ammonia oxidizing bacteria. There are two pathways for nitrous oxide production by ammonia oxidizing bacteria. Either it is produced by the reduction of nitrite (NO_2^-), the so-called nitrifier denitrification pathway, or it is produced as a result of the oxidation of hydroxylamine (NH_2OH) or intermediates of the ammonia oxidation process. The only biological way in which nitrous oxide can be consumed, is by reduction during the denitrification process. Besides these biological pathways, nitrous oxide is also produced during a number of chemical reactions involving nitrite, hydroxylamine and nitroxyl (HNO). The effects of WWTP design and operation on the production and emission of nitrous oxide have been reviewed extensively by Kampschreur et al. (2009), Desloover et al. (2012), Law et al. (2012b) and Wunderlin et al. (2012).

The present study reports nitrous oxide emission data from a 16-month, online monitoring campaign. The objective of this study is to confront the dynamic behaviour of the emission with the process conditions mentioned in literature in order to verify to what extent commonly measured process variables can explain the diurnal and seasonal trend of the nitrous oxide emission. The extent of the dataset described in this study is unprecedented and as a reference dataset it could well serve purposes beyond the scope of the present study, such as calibration and validation of mathematical models and to verify future new hypotheses. All data are therefore made available as supplementary material.

Table 1
Emission factors reported in peer-reviewed full-scale studies.

Reference	Emission factor (N_2O-N as percentage of incoming TKN)
Czepiel et al. (1995)	0.035
Wicht and Beier (1995)	0–14.6 (average 0.6%)
Sümer et al. (1995)	0.001
Sommer et al. (1998)	0.02
Kimochi et al. (1998)	0.01–0.08
Ahn et al. (2010)	0.01–1.8
Foley et al. (2010)	0.006–25.3 [a] (average 0.035 ± 0.027)
Aboobakar et al. (2013)	0.077–0.217
Yoshida et al. (2014)	0.15–4.27
Mikola et al. (2014)	0.02–2.6

[a] As percentage of amount of denitrified nitrogen.

2. Materials and methods

2.1. Field site

The monitoring campaign was performed at Kralingseveer wastewater treatment plant, located near Rotterdam, the Netherlands (51° 54′ 30″ N 4° 32′ 35″ E). Under dry weather conditions the plant treats about 80.000 $m^3\ d^{-1}$ of domestic wastewater. A comprehensive description of the plant's lay-out can be found in Daelman et al. (2012). Basically, the plant consists of a plug flow reactor in series with two parallel Carrousel reactors (Fig. 1). The wastewater first goes through a primary settling tank and an anaerobic selector tank (4800 m^3), where it is mixed with part of the return sludge from the secondary settlers. Then the mixed liquor enters the plug flow reactor, where the complementary part of the return sludge is added. In the plug flow reactor, the mixed liquor passes first through a non-aerated, anoxic zone for denitrification (3600 m^3), followed by an aerated zone (subsurface aeration) for nitrification (8000 m^3). From the aerated zone, about three quarters of the mixed liquor is recycled to the anoxic zone, while the remainder passes on to the two parallel Carrousel reactors (2 × 13.750 m^3), each aerated with three surface aerators. The surface aerators are positioned such that aerated zones are alternated with anoxic zones, to allow for denitrification. Even with all aerators running, each Carrousel reactor has an anoxic zone as the mixed liquor passes from the inlet to aerator #3. The surface aerators are controlled by the ammonia concentration in the Carrousel reactor. Surface aerator #1 operates on/off with a set point of 1.2 g NH_4^+ — N m^{-3}. Aerators #2 and #3 are always running to keep the solids from settling, but they gear up when the ammonia concentration hits 0.6 and 0.9 g NH_4^+ — N m^{-3}, respectively. After passing through the Carrousel reactors, the mixed liquor flows to the secondary settlers, from which part of the sludge is recycled to the selector and to the anoxic zone of the plug flow reactor, and another part is waste sludge. The underflow of the primary settler and the waste sludge are thickened and treated anaerobically. The digestate is dewatered and disposed for incineration. The overflow of the sludge thickening and the reject water from the digestate dewatering are recycled to the primary settler.

2.2. Influent characteristics and plant performance

During the monitoring period, the average COD concentration of the plant's influent was 328 g m^{-3}, the average Total Kjeldahl Nitrogen (TKN) concentration was 42 g N m^{-3}, the ammonia concentration was 30 g N m^{-3}, the average total phosphorous concentration is 6 g P m^{-3} and the average solids concentration was 112 g m^{-3}. The plant's average removal efficiencies were 87% for COD and 81% for total nitrogen. Phosphorous is removed biologically with a total phosphorous removal efficiency of 78%. The plant's effluent requirements are 12.0 g total N–m^{-3}, 2.5 g P m^{-3} and 125 g COD m^{-3}. The mixed liquor suspended solids (MLSS) of the plant is controlled by the operators depending on temperature. As a consequence, the solids retention time (SRT) is the result of the amount of sludge that is wasted in order to maintain the target MLSS.

2.3. Nitrous oxide monitoring

The plug flow reactor and the two Carrousel reactors are covered. The off-gas from these reactors is sent to an ozone washer for disinfection. Because the pressure in the headspace of the reactors is always lower than the atmospheric pressure, all off-gas is collected in the ozone washer. From the ducts leading the off-gas to the ozone washer, gas was pumped continuously to a Servomex infrared gas analyser that measured the concentration of nitrous oxide. The drift of the zero measurement was 1 ppm per week and the drift of the span measurement was 1 ppm or 2% of the reading per week, whichever is the larger.

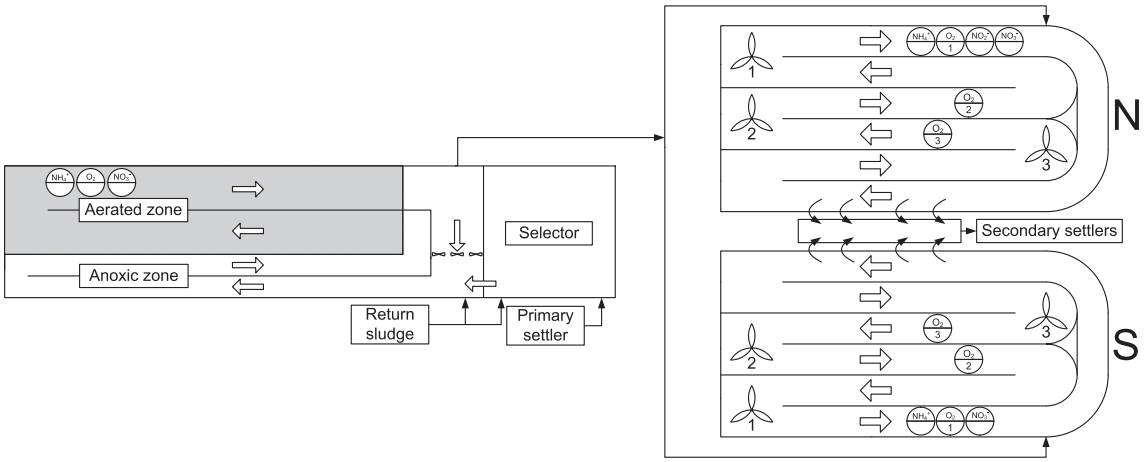

Fig. 1. Lay-out of the plug flow reactor and the two parallel Carrousel reactors, with sensors and aerators indicated. The southern Carrousel reactor mirrors the northern, except for the nitrite sensor which was only available in the northern reactor.

About once a week, the analyser was calibrated with a span gas. To obtain the mass flow rate of nitrous oxide in the pipes, this concentration was multiplied by the constant volumetric gas flow rate (verified with a Testo 435 hot wire anemometer). The relative error on the maximal value of the total emission is 5.4% (see supplementary materials for the calculation). The gas residence time in the headspace of the reactors was lower than 10 min. Given the small volume of the ventilation ducts in comparison with the headspace, the response time of the analyser upon a concentration change in the off-gas was similar to the gas residence time. The configuration of the activated sludge system and the lay-out of the ventilation system allowed establishing the separate nitrous oxide mass flow rates from the plug flow reactor and the two parallel Carrousel reactors. The monitoring protocol is added as supplementary material. The monitoring period covered 16 months, from October 14, 2010 to January 26, 2012, with a month-long interruption in October 2011 due to a technical failure.

2.4. Process data

Dissolved oxygen concentration, nitrate and ammonium concentrations, mixed liquor temperature, aeration rates, total suspended solids concentration in the activated sludge tanks and influent flow rate were available from the plant's SCADA system at 10 minute intervals.

The ammonium loading rate the Carrousel reactor was calculated as follows. The volumetric flow rate from the plug flow reactor to the Carrousel reactor comprises the volumetric influent flow rate and the volumetric flow rate of return sludge (1.2 times the influent flow rate), given that water cannot accumulate in the primary settling tank and the plug flow reactor. This total volumetric flow rate was multiplied by the ammonium concentration measured by the ammonium probe in the aerated zone (see Fig. 1).

For the online nitrite measurements, an S::CAN spectro::lyzer™ probe was used in the northern Carrousel reactor. The linear calibration curve for this sensor used data pairs of spectral signals and nitrite concentration in mixed liquor samples. The nitrite samples for calibration were taken weekly and analysed immediately using Hach Lange kits. The actual calibration of the sensor was performed by the manufacturer.

The Water Board provided laboratory data for plant influent and effluent quality, and for the wastewater quality at the outflow of the primary settler. These data resulted from off-site lab-analyses of volume proportional 24 h composite samples. All process data are provided in the supplementary material.

3. Results and discussion

3.1. Seasonal variability

3.1.1. Magnitude of the emission

Over the entire period, 2.8% of the nitrogen entering the plant was emitted as nitrous oxide. As can be seen in Fig. 2, the emission exhibits a pronounced long-term variability: in the beginning of the monitoring period there was a period with no emission, in December 2010 the emission started to increase, it peaked in March 2011 at about 450 kg N_2O-N d^{-1}. On 31 March 2011, about 11% of the nitrogen entering the plant was emitted as nitrous oxide. From April onwards, the emission gradually decreased again to below 100 kg N_2O-N d^{-1} in summer 2011. In December 2011 there was another short period without any emission at all. In general, the Carrousel reactor (Fig. 2C) appeared to contribute more to the plant's emission than the plug flow reactor (Fig. 2B).

At 2.8% of the incoming nitrogen, the nitrous oxide emission from Kralingseveer WWTP is higher than most values reported by other full-scale studies (Table 1). Yet, such comparisons are delicate since the present study is the first to report long-term, online data, while the others report results from short-term monitoring or low-frequency grab sampling measurements. As discussed in Daelman et al. (2013a), the sampling strategy has a profound effect on the accuracy of the emission estimate. Furthermore, the present study is the only one to monitor the off-gases from the entire liquid surface of covered activated sludge tanks. This allowed for an accurate estimate of the complete amount of nitrous oxide that is emitted from the reactors to the atmosphere. In contrast, Yoshida et al. (2014) used emission plume measurements from an open plant, while all the other studies mentioned in Table 1 used floating hoods. These floating hoods only measure the emission from a small surface and those measurements may therefore not be representative of the total emission of an entire reactor, given the emission's spatial variability.

3.1.2. External disturbances: influent and temperature

The external disturbances to which a wastewater treatment plant is subjected are atmospheric temperature on the one hand and composition, flow rate and temperature of the incoming wastewater on the other hand. Both atmospheric temperature and influent temperature affect the temperature of the mixed liquor. At Kralingseveer WWTP, the influent is mixed with the reject water from the anaerobic sludge digestion before it passes through a primary settler. The influent of the activated sludge system is therefore the effluent

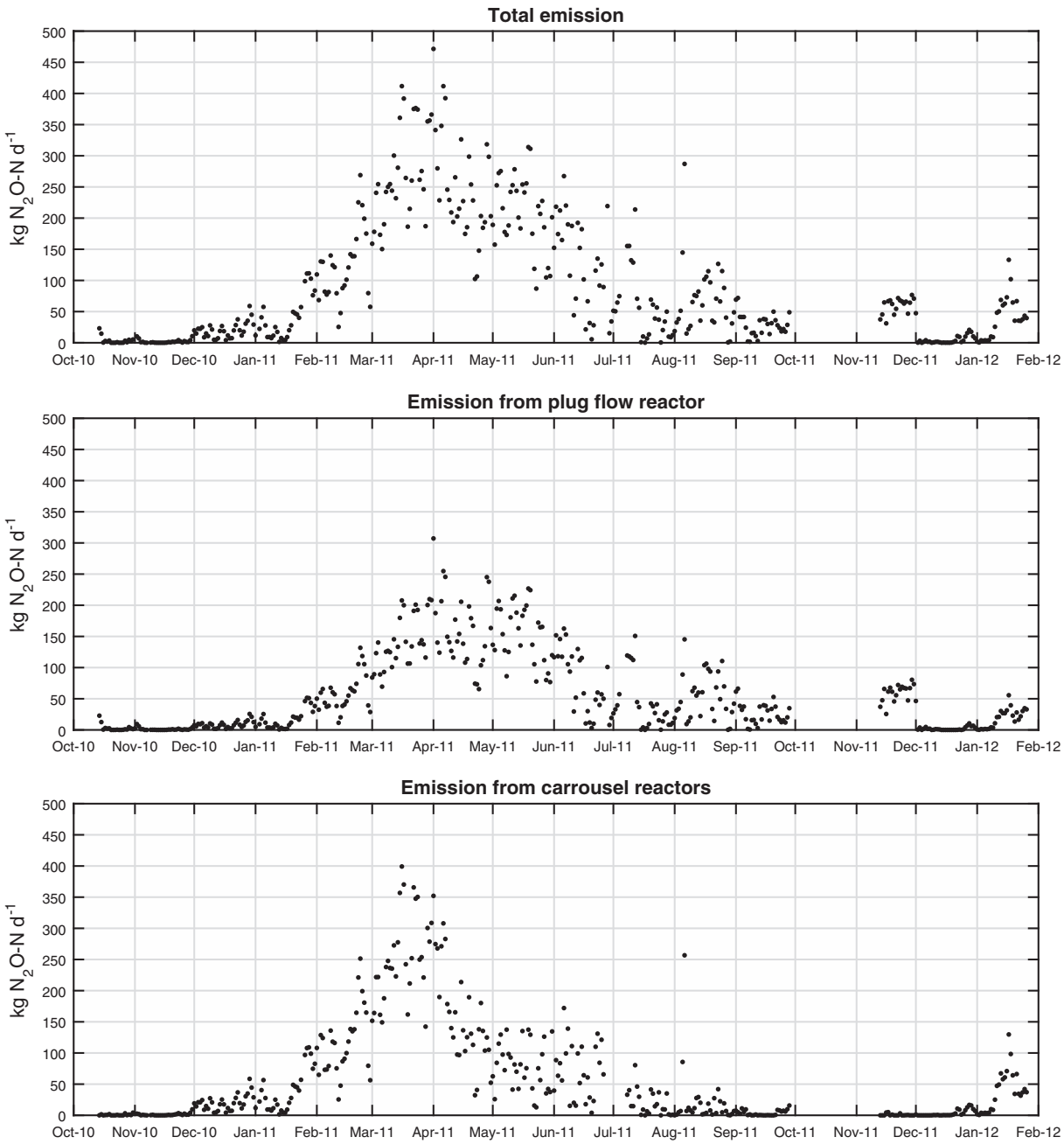

Fig. 2. Daily nitrous oxide emission from the entire plant and from the plug flow reactor and the Carrousel reactor separately. The gap in October/November 2011 is due to equipment failure.

of the primary settler. Pearson's correlation coefficients between these variables and the total emission from the plant are given in Table 2.

Table 2
Pearson's correlation coefficient r and coefficient of determination r^2 between the plant's total daily emission kg N_2O-N d^{-1} on the one hand, and external disturbance variables on the other hand.

	Unit	r	r^2	p-Value
COD concentration of settled wastewater	kg COD m^{-3}	0.22	0.05	0.07
TKN concentration of settled wastewater	kg TKN m^{-3}	0.55	0.30	0.00
COD loading rate from primary settler	kg COD d^{-1}	−0.14	0.02	0.26
TKN loading rate from primary settler	kg TKN d^{-1}	−0.16	0.03	0.19
COD/N at weir of primary settler	–	−0.15	0.02	0.22
Influent flow rate	m^3 d^{-1}	−0.40	0.16	0.00
Mixed liquor temperature	°C	0.09	0.01	0.06

None of the investigated wastewater characteristics correlated strongly with the plant's total nitrous oxide emission. The weak and moderate positive correlations with COD and TKN concentration, respectively, of the settled wastewater and the moderate negative correlation with the influent flow rate may indicate that a more diluted wastewater induces the plant's total emission of nitrous oxide. Yet, as those correlations are rather weak, they may be merely stochastic.

The long-term variability of the emission could not be attributed to any seasonal trend in the COD/N ratio that would influence the denitrification performance (Itokawa et al., 2001; Schulthess, 1995) and neither did the emission correlate with the nitrogen loading rate, as has been suggested in previous research (Burgess et al., 2002a; Chandran et al., 2011; Lotito et al., 2012).

The emission of nitrous oxide from the entire plant did not correlate with mixed liquor temperature. In literature, however, the production of nitrous oxide has been linked to temperature, both high and low.

Ahn et al. (2010) expect the emission of nitrous oxide from plants that are designed for complete nitrogen removal to be higher at higher temperatures because of the higher overall kinetics of the nitrogen transformations. Nonetheless, in a previous study at Kralingseveer WWTP, the opposite was observed (STOWA, 2010). The plant's emission of nitrous oxide was monitored during one week in October 2008 and another week in February 2009. In October 2008, when the water temperature was ca. 18 °C, the emission amounted to only 0.040% of the incoming nitrogen, while it went up to 6.1% in February 2009, when the temperature was as low as 9 °C. Higher emission during colder periods could possibly be explained by increased nitrite concentrations. In WWTPs, nitrite is known to accumulate under low temperatures (Alleman, 1985; Philips et al., 2002; Randall and Buth, 1984). Indeed, at Kralingseveer WWTP there was a strong negative correlation ($r = -0.67$) between the temperature of the mixed liquor and the daily averaged nitrite concentration, and a moderate correlation ($r = -0.50$) between temperature and the daily maximum nitrite concentration in the Carrousel reactor. High nitrite concentrations are commonly recognised as an inducing factor for the production and emission of nitrous oxide (Kampschreur et al., 2009; Wunderlin et al., 2012). The relationship between the emission and the nitrite concentration at Kralingseveer WWTP is discussed in Section 3.1.4 below.

3.1.3. Plant performance and operation

As far as the plant's removal efficiencies are concerned, the removal efficiencies of TKN and total N appeared to correlate only weakly or moderately, respectively, with the plant's total emission. A number of researchers suggested to use the emission of nitrous oxide as an early warning for nitrification process failure (Burgess et al., 2002a,b; Butler et al., 2009) or to control biological ammonia oxidation (Sivret et al., 2008; Wunderlin et al., 2013). Yet, the TKN removal efficiency of Kralingseveer WWTP remained adequate and rather constant throughout the entire monitoring period, which explains its weak correlation with the plant's emission (Table 3). This contradicts any claims that link failing nitrification with the emission of nitrous oxide.

As far as SRT is concerned, there was a strong positive correlation with the emission. This contradicts literature reports suggesting that shorter sludge retention times (SRT) induce the production and emission of nitrous oxide (Lotito et al., 2012; Noda et al., 2003). Wunderlin et al. (2012) suggest maintaining a sufficiently high SRT to avoid accumulation of ammonia and nitrite, but in the present study SRT was not correlated with either nitrite or ammonia.

3.1.4. Conditions in the reactor

In a Carrousel reactor, the concentrations of the nitrogen species can be assumed to approach perfectly mixed conditions (Foley et al., 2010), which makes it possible to compare these concentrations with the emission measured in the off-gas, i.e. the aggregated emission from the entire liquid surface area of the tank. This is not the case for the oxygen concentration: the use of surface aerators results in spatial oxygen concentration gradients in the Carrousel reactor (Fig. 3). In a plug flow reactor, the concentration of the nitrogen species is subject to steep spatial gradients. Local measurements of nitrogen species in a plug flow reactor may therefore not reflect the circumstances that lead to the nitrous oxide emission as it is measured in the off-gas, because the emission measured in the off-gas is the averaged emission coming from the entire liquid surface of the tank. Furthermore, real-time nitrite data were only available for the northern Carrousel reactor. Therefore, the discussion of the nitrogen species concentration is limited to the Carrousel reactor. Correlations of the emission from the Carrousel reactor with relevant variables are shown in Table 4.

Table 3
Pearson's correlation coefficient r and coefficient of determination r^2 between daily emission kg N_2O-N d^{-1} on the one hand, and plant performance variables on the other hand.

		r	r^2	p-Value
COD removal efficiency	%	−0.14	0.02	0.26
TKN removal efficiency	%	0.36	0.13	0.00
Total N removal efficiency	%	0.52	0.27	0.00
Solids residence time	d	0.76	0.58	0.00

The only variable that had a strong correlation with the emission from the Carrousel reactor was the maximum nitrite concentration in the reactor, while there was a moderate correlation with the daily averaged nitrite concentration. So, the emission is mainly related with nitrite peaks. The relationship between an elevated nitrite concentration and the emission of nitrous oxide has been reported in numerous studies (Kampschreur et al., 2009; Schulthess, 1995; Tallec et al., 2006; Wunderlin et al., 2012). In those cases, the nitrifier denitrification pathway was held responsible for the production of nitrous oxide during denitrification. Still, a high nitrite concentration could also have affected the denitrification process with the production and emission of nitrous oxide as a result (Li et al., 2013; Schulthess, 1996; Zhou and Pijuan, 2008). As mentioned in Section 3.1.2 above, the nitrite concentration was correlated negatively with temperature, but the emission itself did correlate only weakly with temperature. So, other variables may be at play, but the occurrence of nitrite peaks in the aeration tank could be used as an indicator for elevated nitrous oxide emission. On the other hand, the detection of nitrous oxide emissions could be used as an indicator for nitrite concentration peaks, which in its turn implies that the nitrification process is not running optimal (Burgess et al., 2002a,b; Butler et al., 2009). Yet, as mentioned in Section 3.1.4 above, the correlation between TKN removal efficiency and plantwide nitrous oxide emissions was weak.

In previous work, the production of nitrous oxide has also been linked to an increased ammonium concentration combined with an elevated oxygen concentration (Aboobakar et al., 2013; Ahn et al., 2010; Rassamee et al., 2011). Under those conditions, the hydroxylamine oxidation pathway for nitrous oxide production is promoted. However, in the present study, the lack of correlation with the average and maximum ammonia concentration suggests that the seasonal trend of the nitrous oxide emission from the Carrousel reactor may not have been caused by hydroxylamine oxidation.

Besides the concentration of the nitrogen species, also the oxygen concentration has been put forward as an important variable with regard to the production and emission of nitrous oxide. An oxygen concentration that is too low during the nitrification process is supposed to induce the nitrifier denitrification pathway (Kampschreur et al., 2008; Rassamee et al., 2011; Tallec et al., 2008; Wunderlin et al., 2012), while the presence of oxygen during the denitrification process inhibits the reduction of nitrous oxide (Gong et al., 2012; Otte et al., 1996; Wunderlin et al., 2012). Yet, during the entire monitoring period the ammonium setpoints controlling the surface aerators remained the same, so the seasonal variability of the emission cannot be linked to operational changes in aeration regime at Kralingseveer WWTP.

3.2. Diurnal variability

3.2.1. Emission and concentrations of nitrogen species

Figure 4A to E show the diurnal trend of the emission from the northern Carrousel reactor as well as the concentrations of the nitrogen species and ammonium loading rate during one week in January 2012. The supplementary material provides the same data for the entire monitoring period.

Although the absolute value of the nitrous oxide emission varies a lot during the monitoring period, with even some periods without emission at all (e.g. Autumn 2010), the emission from the Carrousel reactor exhibits a pronounced diurnal trend throughout the entire monitoring period. At noon, the nitrous oxide emission starts to rise, to reach a peak around midnight. Then the emission starts to decrease again, reaching a low shortly before noon. Ultimately, this variability can be traced back to the diurnal pattern of the nitrogen loading rate

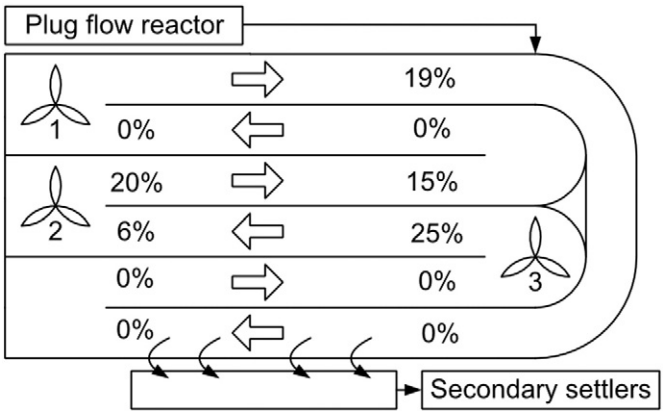

Fig. 3. Dissolved oxygen concentration (% of saturation) at several locations in the northern Carrousel reactor, measured on 20 May 2011. During the measurements, all surface aerators were operational.

to the Carrousel reactor (Fig. 4E), since nitrous oxide is only produced when nitrogen is being converted. The ammonia loading rate to the Carrousel reactor shown in Fig. 4E exhibits the typical pattern of the nitrogen loading rate to a wastewater treatment plant, be it with a time lag that corresponds with the hydraulic residence time in the reactors preceding the Carrousel reactor, i.e. the primary settler, the selector and the plug flow reactor. Using the average dry weather flow rate of the plant, this hydraulic retention time amounted to 4.5 h. This explains why the nitrous oxide emission from the Carrousel reactor peaks at night.

In the present study, the contribution of either nitrification of denitrification to the emission from the tank could not be established, since the off-gas monitoring comprised the composite emission from both the aerobic and anoxic zones in the reactors. The following sections attempt to verify to what extent the observations of the full-scale Carrousel reactor agree with the established production pathways for nitrous oxide, i.e. nitrifier denitrification and hydroxylamine oxidation during nitrification on the one hand and heterotrophic denitrification on the other hand.

As the diurnal variability of the emission is superimposed on the seasonal variability, the long-term online data of the emissions and the process variables (in contrast with the daily averaged data above) were too scattered to obtain meaningful statistical correlations. Yet, when looking at the short-term trends, some clear relationships can be distinguished.

3.2.2. Nitrification

During the depicted period, the ammonium concentration in the Carrousel reactor is rather constant around 1.5 g $NH_4^+ - N\ m^{-3}$ but the ammonium loading rate shows a pronounced diurnal pattern that coincides with the diurnal trend of the nitrous oxide emission. This indicates that the emission peak could be related to a peak in the ammonia oxidation rate, in agreement with Chandran et al. (2011), De Clippeleir et al. (2012), Law et al. (2012a) andSchneider et al. (2013, 2014). Usually, the ammonia oxidation rate in activated sludge processes is modelled by a multiplicative Monod-model as in Eq. (1) (Henze et al., 1987; Ni et al., 2013).

$$\mu = \mu_{max} \cdot \frac{S_{NH_4}}{S_{NH_4} + K_{s,NH_4}} \cdot \frac{S_{O_2}}{S_{O_2} + K_{s,O_2}} \quad (1)$$

The commonly assumed value for the ammonium half-saturation constant K_{s,NH_4} is 1.0 mg $NH_4^+N\ L^{-1}$ (Henze et al., 1987). Given that the ammonium concentration is always well above the ammonium half-saturation constant (Fig. 4D), the Monod term for ammonia remains constant, so the ammonia oxidation rate depends essentially on the oxygen concentration. Indeed, the emission coincides with the aerated periods (Fig. 5). So, the emission of nitrous oxide from the Carrousel reactor could possibly be related to the nitrification rate, which in its turn is governed by the oxygen concentration. The relationship between the ammonia oxidation rate and the nitrous oxide production rate is usually explained by referring to the hydroxylamine pathway (Chandran et al., 2011; Law et al., 2012a). It should be noted, however, that the oxygen concentration in the Carrousel reactor is highly heterogeneous because of the use of surface aerators (Fig. 3). As the mixed liquor passes along the surface aerator, it gets oxygenated, but further on along the flow path, the water gets gradually depleted of oxygen as the latter is consumed in the oxidation processes. Even when all the surface aerators are running, the Carrousel reactor contains anoxic zones to allow for denitrification. As a result, steep oxygen gradients occur in the Carrousel reactor and even when all surface aerators are running, certain reactor zones will have an oxygen concentration in between 0 and 1.5 g $O_2\ m^{-3}$. On top of such large-scale gradients between reactor zones, also small-scale variability exists within the sludge flocs. Since ammonia oxidizing bacteria have a higher oxygen affinity than nitrite oxidizing bacteria, low oxygen concentrations may result in the accumulation of nitrite (Blackburne et al., 2008). In its turn, the combination of a low oxygen concentration with an elevated nitrite concentration has since long been known to induce the nitrifier denitrification pathway (Kampschreur et al., 2008; Kim et al., 2010; Peng et al., 2014; Tallec et al., 2006; Wunderlin et al., 2012). This would explain the agreement between the trend in the nitrite concentration and the trend of the nitrous oxide emission from the Carrousel reactor that can be seen in Fig. 4. So, both nitrification pathways could possibly have contributed to the emission, but the emission's dependence on the nitrite concentrations and the prevalence of low oxygen zones in the reactor suggest that nitrifier denitrification was the dominant pathway for nitrous oxide production by nitrifiers.

Table 4
Pearson's correlation coefficient r and coefficient of determination r^2 between daily emission from the northern Carrousel reactor kg N_2O-N d^{-1} on the one hand, and conditions in the reactor on the other hand.

	r	r^2	p-**Value**
Average nitrite conc.	0.40	0.16	0.00
Max. nitrite conc.	0.73	0.53	0.00
Average nitrate conc.	−0.29	0.08	0.00
Max. nitrate conc.	−0.35	0.12	0.00
Average ammonium conc.	−0.10	0.01	0.04
Max. ammonium conc.	−0.13	0.02	0.01
Temperature	−0.13	0.02	0.01
Solids concentration	−0.06	0.00	0.22

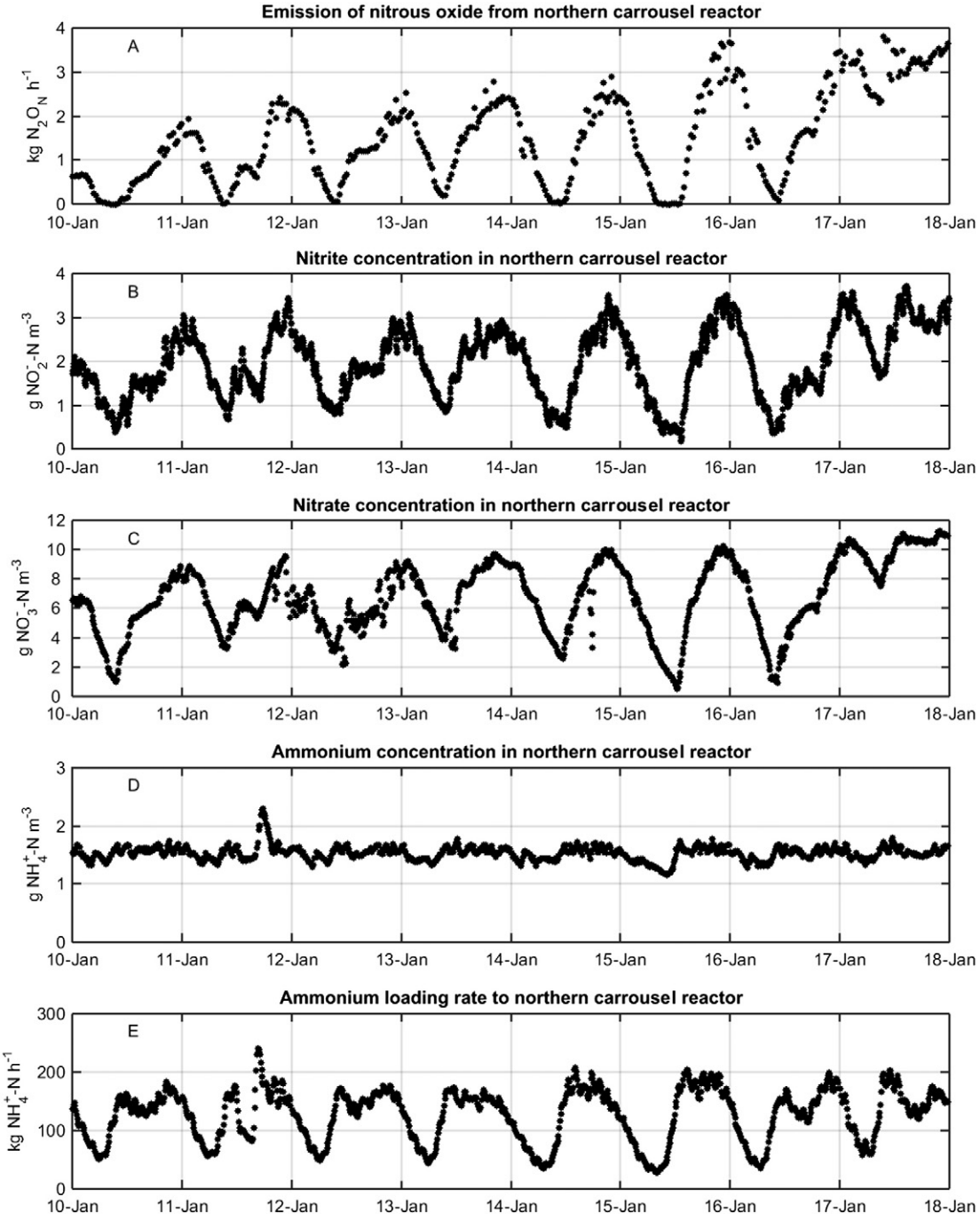

Fig. 4. Diurnal trend of nitrous oxide emission (A), nitrite concentration (B), nitrate concentration (C), ammonium concentration (D) and calculated ammonium loading rate (E) for the northern Carrousel reactor for one week in 2012. Vertical lines indicate midnight.

3.2.3. Denitrification

The possibility that the nitrous oxide emitted from the Carrousel reactor was produced by nitrifiers does not exclude the production of nitrous oxide by heterotrophic denitrifying organisms. The observation that the diurnal emission pattern coincides with the diurnal pattern of the nitrate concentration in the tank indicates that failing denitrification may contribute to the production and emission of nitrous oxide. The accumulation of nitrate indicates that the denitrification rate cannot keep up with the nitrification rate. The observed nitrate accumulation does not imply that no nitrate was reduced at all, however there are several reasons why a stop of the denitrification process at the last step, resulting in the accumulation of nitrous oxide, is likely. In heterotrophs, nitrous oxide reductase could be inhibited by the elevated nitrite concentrations in the reactor (Betlach and Tiedje, 1981; Li et al., 2013; Schulthess, 1996; Zhou and Pijuan, 2008). Also the lack of electron donor has been mentioned as a possible cause for nitrous oxide emissions (Chung and Chung, 2000; Hanaki et al., 1992; Itokawa et al., 2001; Schulthess, 1995), yet the COD/N ratio of the stream entering the Carrousel tank was not measured in this study. Finally, insufficient anoxia may result in incomplete denitrification or even no denitrification at all. Of all the enzymes involved in denitrification, nitrous oxide reductase, the enzyme that catalyses the reduction of nitrous oxide to nitrogen gas, was found to be the most sensitive to oxygen in *Pseudomonas nautica* (Bonin et al., 1989). Denitrification in the presence of about

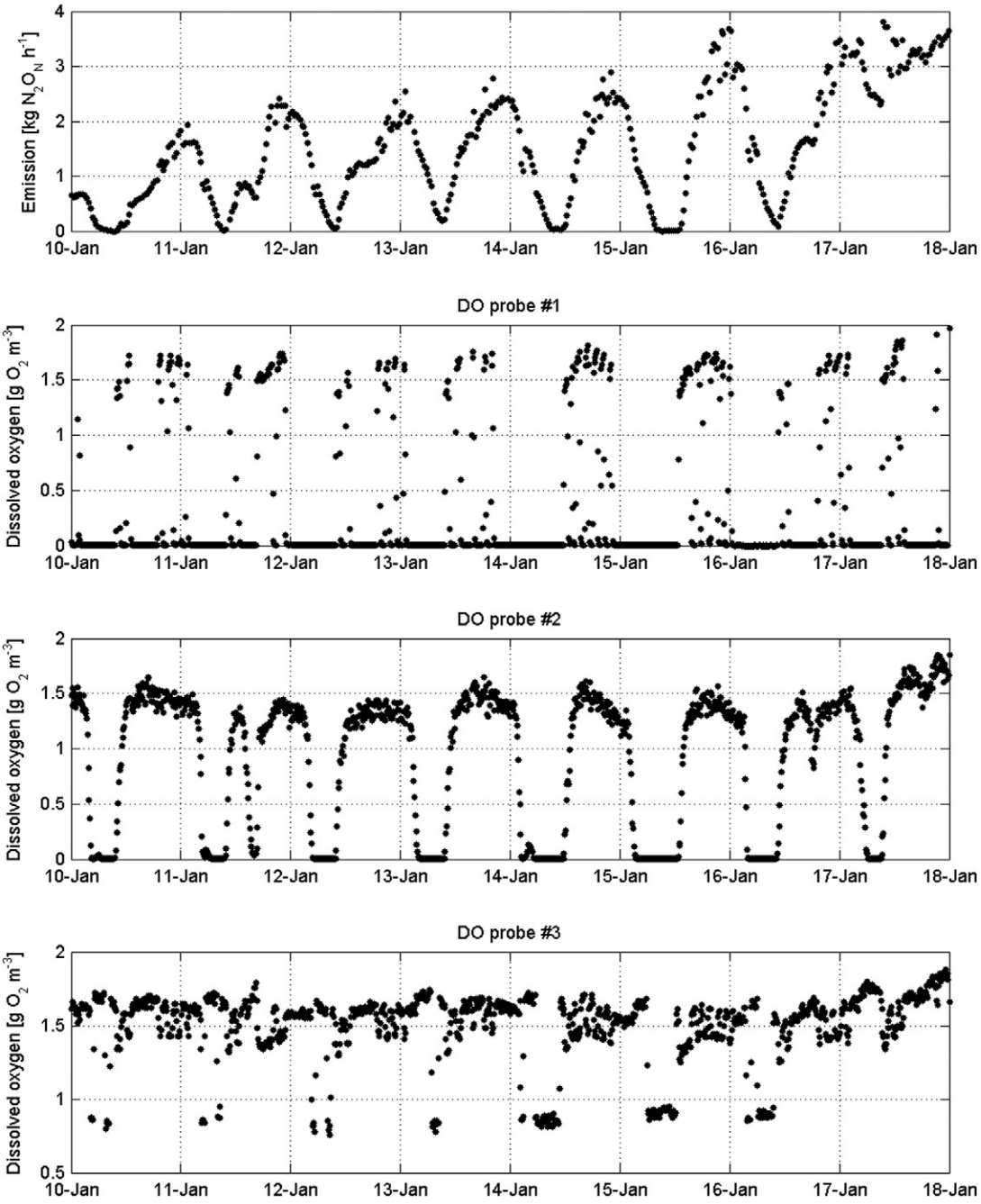

Fig. 5. Dissolved oxygen concentrations in northern Carrousel reactor.

0.3 g O_2 m^{-3} can already lead to significant nitrous oxide emission (Tallec et al., 2008). The Carrousel reactor may indeed be characterized by insufficient anoxia. The diurnal nitrate concentration is in step with the activity of the aerators, which indicates that the denitrification process in the Carrousel reactor is hampered by the aeration. Still, the possible production of nitrous oxide as a result of incomplete denitrification does not rule out the production of nitrous oxide during the nitrification process. Yet, even if the nitrous oxide is produced during nitrification, insufficient denitrification would still contribute to the emission since nitrous oxide is not or to a lesser extent consumed.

3.2.4. Coupling of production and emission

One could argue that the diurnal trend of the emission is caused by stripping and that the emission does not reflect the production of nitrous oxide. Yet, if emission and production were uncoupled, most of the produced nitrous oxide would remain dissolved in the liquid phase, and it would be stripped as soon as the aerators start working. In that case, the emission would exhibit a steep increase as soon as the aerators are switched on, followed by an exponential decrease as the liquid gets depleted of nitrous oxide (Mampaey et al., 2015). This emission profile cannot be seen in Fig. 6. The gradual increase of the emission after the surface aerators start working indicates that the emission is not merely due to stripping of previously accumulated nitrous oxide. Only the sharp peaks on top of the wider diurnal peaks could be attributed to the fast on/off behaviour of the aerator #1.

3.3. Towards mitigation

The analysis of both the seasonal and the diurnal data from this long-term full-scale monitoring campaign confirmed earlier reports stating

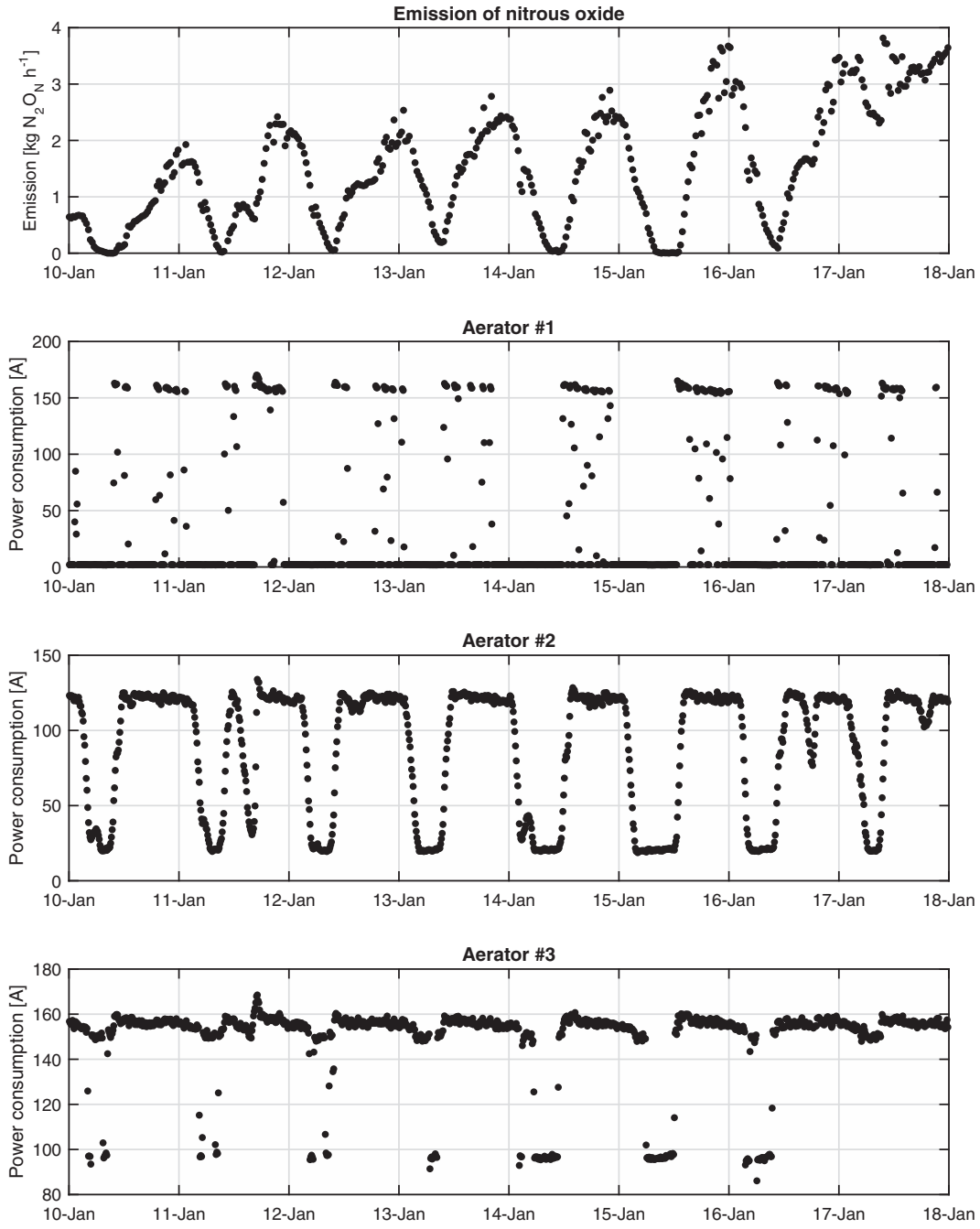

Fig. 6. Emission with power consumption of the northern carrousel's surface aerators.

that peaks in the nitrite concentration are an important trigger for the emission of nitrous oxide emission (Desloover et al., 2012; Kampschreur et al., 2009). Other commonly available operational data of the WWTP could not explain the long-term trend of the emission. The analysis of the diurnal variability in particular suggests that nitrifier denitrification and heterotrophic denitrification are the most probable pathways for the production of nitrous oxide in this WWTP. In the Carrousel reactors, both pathways benefit from the prevalence of suboptimal oxygen concentrations, caused by the use of surface aerators. The use of surface aerators results in reactor zones where the oxygen concentration is too low to allow for complete nitrification, resulting in the accumulation of nitrite. The combination of elevated nitrite concentrations and low oxygen concentrations is known to induce the nitrifier denitrification pathway. On the other hand, the reactor appears to lack sufficient anoxic space to allow for proper denitrification. This could explain part of the nitrous oxide production, but it also prevents consumption of nitrous oxide, as denitrification is the only biological way to reduce nitrous oxide. Also the plug flow reactor of this WWTP is characterized by very heterogeneous oxygen concentrations since about three quarters of the mixed liquor is recycled from the aerated zone to the anoxic zone. Instead of using reactors with mixed oxygen concentrations, it may be better to use dedicated reactors in which the aerobic and anoxic conditions can be optimized for nitrification and denitrification, respectively, avoiding oxygen concentrations that are suboptimal for both processes.

It should be noted that some variables that were investigated in literature could not be accounted for in the present monitoring campaign. Among those variables are storage compounds (Jia et al., 2012; Lemaire et al., 2006; Zeng et al., 2003), iron (Butler and Gordon, 1986; Kampschreur et al., 2011) or inorganic carbon (Peng et al., 2015). Neither did this study cover the effects of microbial community

dynamics or microbial adaptation on the production of nitrous oxide as it occurred in the work by van Benthum et al. (1998) and Chandran et al. (2011). The verification of this work on full-scale conditions may help closing the knowledge gap that still stands in the way of a proper mitigation strategy.

4. Conclusion

- This study exposed the different time scales of the dynamic behaviour of the nitrous oxide emission: a pronounced diurnal variability was superimposed on a seasonal trend.
- The diurnal trend of the emission coincided with the diurnal trends of the nitrite and nitrate concentration and with the ammonia loading rate.
- The seasonal trend of the emission correlated with the daily maximum nitrite concentration. The occurrence of daily nitrite peaks could therefore be used as a diagnostic for the emission of nitrous oxide, while the detection of nitrous oxide emission indicates the occurrence of nitrite concentration peaks.
- The dataset presented in this manuscript suggests that nitrous oxide is mainly produced as a result of sub-optimal oxygen concentrations leading to denitrification by nitrifying bacteria and a hampered denitrification process.

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Appendix A. Supplementary data

For the entire monitoring period, all data that were presented in this paper are available in the supplementary materials. These long-term, real-time data of a covered wastewater treatment plant are scarce, and the authors encourage other researchers to use these data, e.g. for calibrating and validating models. Supplementary data to this article can be found online at http://dx.doi.org/10.1016/j.scitotenv.2015.06.122.

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