Interlinkages between operational conditions and direct and indirect greenhouse gas emissions in a moving bed membrane biofilm reactor

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Abstract
Nitrous oxide (N_2O) can be emitted during wastewater treatment contributing to the global warming due to its high global warming potential,. During the last ten years, several efforts have been provided to improve knowledge on: key mechanisms, operating factors and influent features affecting the N_2O production/emission. However, the knowledge on the investigated issues is not completely mature. Indeed, in terms of mathematical modelling, literature shows that a reliable model has not yet been established due to the huge data set required and the complexity of the mechanistic models indicated as the most accurate. In this work, the first attempt to perform a multiregression analysis is presented with the final aim to get a simple and easy tool for N_2O estimation from wastewater treatment plant. The multiregression analysis has been performed by testing both simple and complex equations by means of Monte Carlo simulations. Data acquired from an University Cape Town moving bed membrane bioreactor pilot plant have been adopted. The pilot plant has been operated at different sludge retention times. Results of the simple linear regression analysis show that such approaches are suitable to predict N_2O flux emitted from each tank of the plant and dissolved in the permeate. For some tested cases, a high efficiency (obtained comparing simulated and measured data) was obtained (e.g., 0.96 for N_2O-N dissolved in the effluent). The results show that the dependence with the available measured data changes with the operational conditions. Conversely, results related to the complex multiregression analysis reveal that no unique equation valid for different operational conditions can be established.

Keywords
GHG; regression analysis; wastewater; N_2O; pilot plant; nutrients.

INTRODUCTION
Wastewater treatment entails direct emissions of greenhouse gases (GHGs), such as nitrous oxide (N_2O), as well as indirect emissions resulting from power requirements (Flores-Alsina et al., 2014).
Among the possible GHGs produced by a wastewater treatment plant (WWTP) (e.g., CO_2, CH_4 and N_2O) N_2O merits particular interest due to its great climate change potential. Indeed, the global warming potential (GWP) of N_2O is 298 times higher than carbon dioxide (CO_2), moreover it has the capability to react with stratospheric ozone causing ozone layer depletion (IPCC, 2013). N_2O is mainly produced in biological nitrogen removal (BNR) both via nitrification and denitrification processes (Kampschreur et al., 2009).
Process operations aimed at the reduction of N_2O could conflict with the effluent quality, for example the reduction of the aeration flow rates to decrease the energy requirements could negatively affect the biological process due to the dissolved oxygen (DO) limitation. Moreover, the oxygen limitation could favour the N_2O production increasing the mass of discharged pollutants (Guo et al., 2016). Therefore, the identification of GHG mitigation strategies as trade-off between operational costs and effluent quality index is a very ambitious challenge. With this regard, during the last years several efforts have been performed in order to better understand the key mechanisms surrounding around the N_2O formation/emission (Kampschreur et al., 2009; Law et al., 2012) and to identify key factors mainly affecting its formation. Despite researchers seem to converge on the ammonia oxidation biomass (AOB) denitrification as the predominant process responsible for N_2O emission, the interrelationship with the incomplete oxidation of hydroxylamine and heterotrophic denitrification is still poorly known (Wunderlin et al., 2012). Further, the literature suggests that in processes aimed at simultaneous nitrogen and phosphorous removal, the role of polyphosphate

accumulating organisms (PAOs) in the production of N_2O cannot be neglected (e.g. Wang et al., 2011; Zhou et al., 2012). Several studies have also demonstrated that the N_2O production is strongly depending on the plant operating conditions and on the influent wastewater features (Kampschreur et al., 2009; Peng et al., 2015). Therefore, a huge variations of N_2O emissions can be obtained among different WWTPs and inside the same plant due to the different features of the influent wastewater over the day (dynamic conditions). Such a condition, coupled with the poor knowledge on the key processes, have involved several difficulties in establishing an accurate mathematical model able to predict the N_2O emitted from WWTPs (Mannina et al., 2016a). Indeed, despite several mathematical models have been proposed, tested and compared in literature (Corominas et al., 2012; Ni et al., 2013; Spérandio et al., 2016) a reliable model has not yet been established. Furthermore, the existing models have often the limit to be very complex (hundreds of involved model factors and modelled processes) and require high computational costs . Therefore, the need of establishing simple interrelationships, feasible to be used even from operators, among operational conditions/influent features/effluent quality/available monitoring data and the emitted N_2O is required.
The paper presents a multivariate analysis performed by adopting an extensive dataset acquired during the monitoring of a University Cape Town (UCT) moving bed (MB) membrane bioreactor (MBR) pilot plant. The pilot plant was operated by adopting different operational conditions in terms of sludge retention time (SRT). The main aim of the work is to establish a simple mathematical model able to explain the interlinkage existing among the operational conditions/influent features/effluent quality/available monitoring data and the emitted N_2O. The study represents a first attempt to provide useful and (easy to be used) tools to the plant operators to quantify the N_2O emitted by the WWTP.

MATERIALS AND METHODS

The pilot plant and operational conditions

The UCT-MB-MBR pilot plant (Figure 1) consisted of an anaerobic, an anoxic and an aerobic in-series tank according to the UCT scheme (Ekama et al., 1983).

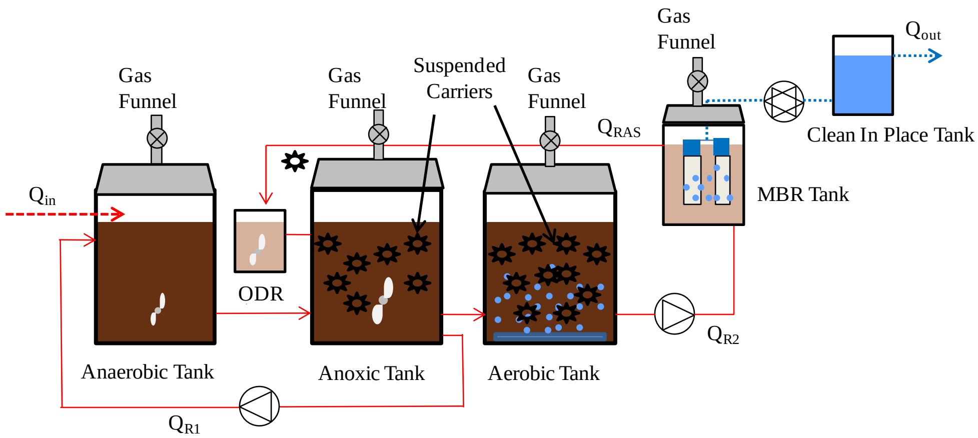

Figure 1. Schematic lay-out of the UCT-MBMBR pilot plant (Mannina et al., 2016b).

The solid-liquid separation phase was achieved by means of an ultrafiltration hollow fibre membrane module (PURON®), located inside a dedicated aerated tank (MBR tank). The membrane was periodically backwashed (every 9 min for a period of 1 min) by pumping, from the Clean In Place (CIP) tank a volume of permeate back through the membrane module. An oxygen depletion reactor (ODR) allowed the oxygen stripping/consumption in the mixed liquor recycled from the MBR tank to the anoxic one (Q_{RAS}). The pilot plant contained plastic carriers in the anoxic and aerobic tanks with filling ratio of 15% and 40%, respectively. Each tank was equipped with a

specific cover that enabled to capture the N_2O produced from each tank as well as from the entire pilot plant. Each tank of the pilot plant is equipped with a separate cover that enabled the capture of the N_2O produced from the tank as well as from the entire pilot plant.
The UCT-MB-MBR pilot plant was operated for almost 150 days and was fed with a mixture of real domestic and synthetic wastewater. During the plant operation three different experimental phases each characterized by a different SRT value were established: i. Phase I, with SRT = ∞; ii. Phase II, with SRT = 30 days; iii. Phase III, with SRT = 15 days. The extraction flow rate was set equal to 20 L h^{-1} (Q_{IN}). During the pilot plant operations, a 20 L h^{-1} flow rate (Q_{R1}) was continuously recycled from the anoxic to the anaerobic tank. Furthermore, a 100 L h-1 flow rate (Q_{R2}) of mixed liquor was pumped from the aerobic to the MBR tank. A net permeate flow rate of 20 L h^{-1} was extracted (Q_{OUT}) through the membrane module. The recycled activated sludge (Q_{RAS}) from the MBR to the anoxic tank through the ODR compartment was equal to 80 L h^{-1}.

Experimental campaign
During pilot plant operations, the influent wastewater, the mixed liquor inside the anaerobic, anoxic, aerobic and MBR tank and the effluent permeate have been sampled and analysed for TSS, volatile suspended solids (VSS), total chemical oxygen demand (COD_{TOT}), supernatant COD (COD_{SUP}), ammonium nitrogen (NH_4-N), nitrite nitrogen (NO_2-N), nitrate nitrogen (NO_3- N), total nitrogen (TN), phosphate (PO_4-P), total phosphorus (TP). All analyses were carried out according to the Standard Methods (APHA, 2005); pH, dissolved oxygen (DO) and temperature were also monitored in each tank by using a multi-parameter probe. Samples of carriers were also withdrawn in order to evaluate the concentration of the attached biomass (i.e., biofilm).
During the pilot plant operation, liquid and gaseous samples were withdrawn from the anaerobic, anoxic, aerobic and MBR tanks and analysed to determine the N_2O-N concentration. Dissolved gas sampling was conducted on the basis of the head space gas method derived from Kimochi et al. (1998). N_2O-N concentration was measured by using a Gas Chromatograph (Thermo Scientific™ TRACE GC) equipped with an Electron Capture Detector. Furthermore, the N_2O-N fluxes (gN_2O-N m^{-2} h^{-1}) from all the tanks were quantified by measuring the gas flow rates, Q_{gas} (L min^{-1}) and adopting the N_2O-N concentration measured in the gas samples.

Multiregression analysis
The multiregression has been performed in order to point out general relationships for the N_2O-N produced in a WWTP and the plant operation conditions or the available measured data. Two different types of analysis have been performed: a simple linear regression analysis and a complex regression analysis.

Simple regressions. A simple linear equation (Equation 1) (LINs) has been tested in order to find a relationship between the dependent variable (Y) and the independent variable (X).

$$Y = c_1 \cdot X_1 + c_2 \qquad [1]$$

where c_1 and c_2 are the regression coefficients.
The simple multiregression analysis has been performed by considering the following dependent variables: N_2O-N flux emitted from the anaerobic tank (N_2O-N $flux_{ANAER}$), N_2O-N flux emitted from the anoxic tank (N_2O-N $flux_{ANOX}$), N_2O-N flux emitted from the aerobic tank (N_2O-N $flux_{AER}$), N_2O-N flux emitted from the MBR tank (N_2O-N $flux_{MBR}$) and the N_2O-N permeate dissolved concentration (N_2O-N $dissolved_{OUT}$). The independent variables summarized in Table 1 have been taken into account.

Complex regressions. Three complex equations have been tested in order to find a relationship between the dependent variable (Y) and the independent variable (X): multiple linear (LINm) (Equation 2), multiple exponential (EXP) (Equation 3) and sum of exponential (SumEXP) (Equation 4).

$$Y = c_1 \cdot X_1 + \cdots + c_n \cdot X_m \quad [2]$$

$$Y = c_1 \cdot X_1^{c2} + \cdots + c_{n-1} \cdot X_m^{cn} \quad [3]$$

$$Y = c_1 \cdot X_1^{c2} \times \ldots \times c_{n-1} \cdot X_m^{cn} \quad [4]$$

where $c_1, \ldots, c_n$ are the regression coefficients and $X_1, \ldots, X_m$ refer to the independent variables.

Table1. Independent variables taken into account during the multiregression analysis; only variables of the grey lines have been considered during the complex analysis. *refer to the weighted sum of the concentrations of each tank.

Symbol	Definition	unit
$COD_{TOT, IN}$	Influent concentration of total COD	mg/L
$N\text{-}NH_{4,IN}$	Influent concentration of ammonia	mg/L
$P_{TOT,IN}$	Influent concentration of total phosphorus	mg/L
$P\text{-}PO_{4,IN}$	Influent concentration of phosphate	mg/L
$N\text{-}NO_{2\_AER}$	Nitrite concentration in the aerobic tank	mg/L
$N\text{-}NO_{2\_ANOX}$	Nitrite concentration in the anoxic tank	mg/L
C/N	Influent carbon nitrogen ratio	-
$COD_{TOT,OUT}$	Permeate concentration of total COD	mg/L
$BOD_{5,OUT}$	Permeate concentration of total BOD	mg/L
$N\text{-}NH_{4,OUT}$	Permeate concentration of ammonia	mg/L
$N\text{-}NO_{3,OUT}$	Permeate concentration of nitrate	mg/L
$NO_2\text{-}N_{,OUT}$	Permeate concentration of nitrite	mg/L
$P\text{-}PO_{4,OUT}$	Permeate concentration of phosphate	mg/L
$\eta COD_{,BIO}$	Biological COD removal efficiency	-
$\eta COD_{,TOT}$	Total COD removal efficiency	-
DO_{AER}	Dissolved oxygen concentration in the aerobic tank	mg/L
DO_{ANOX}	Dissolved oxygen concentration in the anoxic tank	mg/L
DO_{MBR}	Dissolved oxygen concentration in the MBR tank	mg/L
pH_{AER}	pH in the aerobic tank	-
pH_{ANOX}	pH in the anoxic tank	-
ηNITR	Nitrification efficiency	-
ηDENIT	Dentrification efficiency	-
ηN_{TOT}	Total nitrogen removal efficiency	-
ηP	Phosphorus removal efficiency	-
TSS	Total suspended solids concentration*	g/L
SRT	Sludge Retantion Time	day
Biofilm	Attached biofilm concentration*	g/L

The complex multiregression analysis has been performed for two dependent variables: the sum of the N_2O-N flux emitted from each tank ($\sum N_2O$-N flux) and the N_2O-N permeate dissolved concentration (N_2O-N dissolved_{OUT}). The independent variables summarized in Table 1 have been take into account.

Numerical settings and details on the multiregression analysis. In order to test each equation (both simple and complex) 10,000 Monte Carlo simulations have been performed by varying the equation coefficients within a wide range. For each simulation, simulated data were compared with the measured data and the Nash and Sutcliffe (1970) efficiency (Equation 5) has been evaluated.

$$Efficiency = 1 - \frac{\sum_{i=1}^{n}\left(Y_{meas,i} - Y_{sim,i}\right)^2}{\sum_{i=1}^{n}\left(Y_{meas,i} - Y_{aver,meas,i}\right)^2} \quad [5]$$

where: $Y_{meas,i}$ represents the measured value of the ith dependent state variable; $Y_{sim,i}$ represents the simulated value of the ith dependent state variable; $Y_{aver,meas,i}$ represents the average of the measured values of the ith dependent state variable.
The Nash and Sutcliffe's efficiency can range from $-\infty$ to 1. For the case in which the modelled data perfectly match the measured ones, the efficiency is equal to 1. The value of the efficiency equal to zero indicates that the modelled data are as accurate as the average of the observed data.
The simple regression analysis has been applied for each of the experimental phase. By adopting knowledge acquired by means of simple regression analysis, the complex regression analysis has been applied by considering all the data of the three experimental phases. During the complex regression analysis, all possible combinations among dependent and independent variables have been tested.

RESULTS AND DISCUSSION

Simple linear regression analysis

Table 2 summarizes the results of the simple linear regression analysis. More precisely, data reported in Table 2 refer to the maximum efficiency obtained for each depended variables under study; the relative independent variable and the values of the equation coefficients (c_1 and c_2) are also reported in Table 2.
By analysing data reported in Table 2 one can observe that, excepting for sporadic cases, for the same dependent variable the maximum efficiency has been obtained considering different independent variables for the three experimental phases. For example, the dependent variable N_2O-N $flux_{AER}$ mainly depends on the DO_{AER} during the Phase I (SRT = ∞) and on the attached biofilm concentration during the Phase III (SRT = 15 days) (Table 2). Such a result has peculiar interest because suggests that by varying the SRT different variables can be adopted to predict the N_2O. For example, in case of low SRT (e.g., 15 days) the results obtained here suggest to monitor the NO_2-N concentration, easier to be measure than N_2O, and to adopt it in order to predict the amount of N_2O discharged with the treated effluent. . By analysing data of Table 2 one can also observe that during the Phase I the N_2O-N flux of the aerobic and anoxic tank (N_2O-N $flux_{AER}$ and N_2O-N $flux_{ANOX}$, respectively) flux is mainly correlated with the NO_2-N_{ANOX} with an efficiency value of 0.52 for both cases (Table 2).

Table 2. Value of the regression coefficients (c_1 and c_2) and of the efficiency related to the maximum efficiency obtained for each investigated depended variables

		Dependent variables				
		N_2O-N $flux_{ANAER}$	N_2O-N $flux_{ANOX}$	N_2O-N $flux_{AER}$	N_2O-N $flux_{MBR}$	N_2O-N $dissolved_{OUT}$
		Unit				
Phase		mg N_2O-N m^{-2} h^{-1}				mg N_2O-N L^{-1}
I	c_1	0.0884	0.1384	12.902	0.226	0.0004
	c_2	-0.302	0.069	1.0072	-5.438	0.0061
	Independent variable	TSS	NO_2-N_{ANOX}	NO_2-N_{ANOX}	NH_4-N_{IN}	NO_3-N_{OUT}
	Efficiency	**0.11**	**0.52**	**0.52**	**0.2**	**0.1**
II	c_1	0.2177	0.2476	3.5283	4.2862	0.0219
	c_2	0.1741	0.1358	-10.958	-201.3	-0.1094
	Independent variable	NO_2-N_{ANOX}	NO_2-N_{ANOX}	DO_{AER}	η_{NITR}	COD_{OUT}
	Efficiency	**0.35**	**0.6**	**0.5**	**0.26**	**0.72**
III	c_1	2.0481	-5.587	-18.15	0.2604	0.0382
	c_2	-15.064	10.055	34.879	6.46	0.0042
	Independent variable	pH_{AER}	Biofilm	Biofilm	PO_4-P_{OUT}	NO_2-N_{AER}
	Efficiency	**0.12**	**0.36**	**0.67**	**0.52**	**0.94**

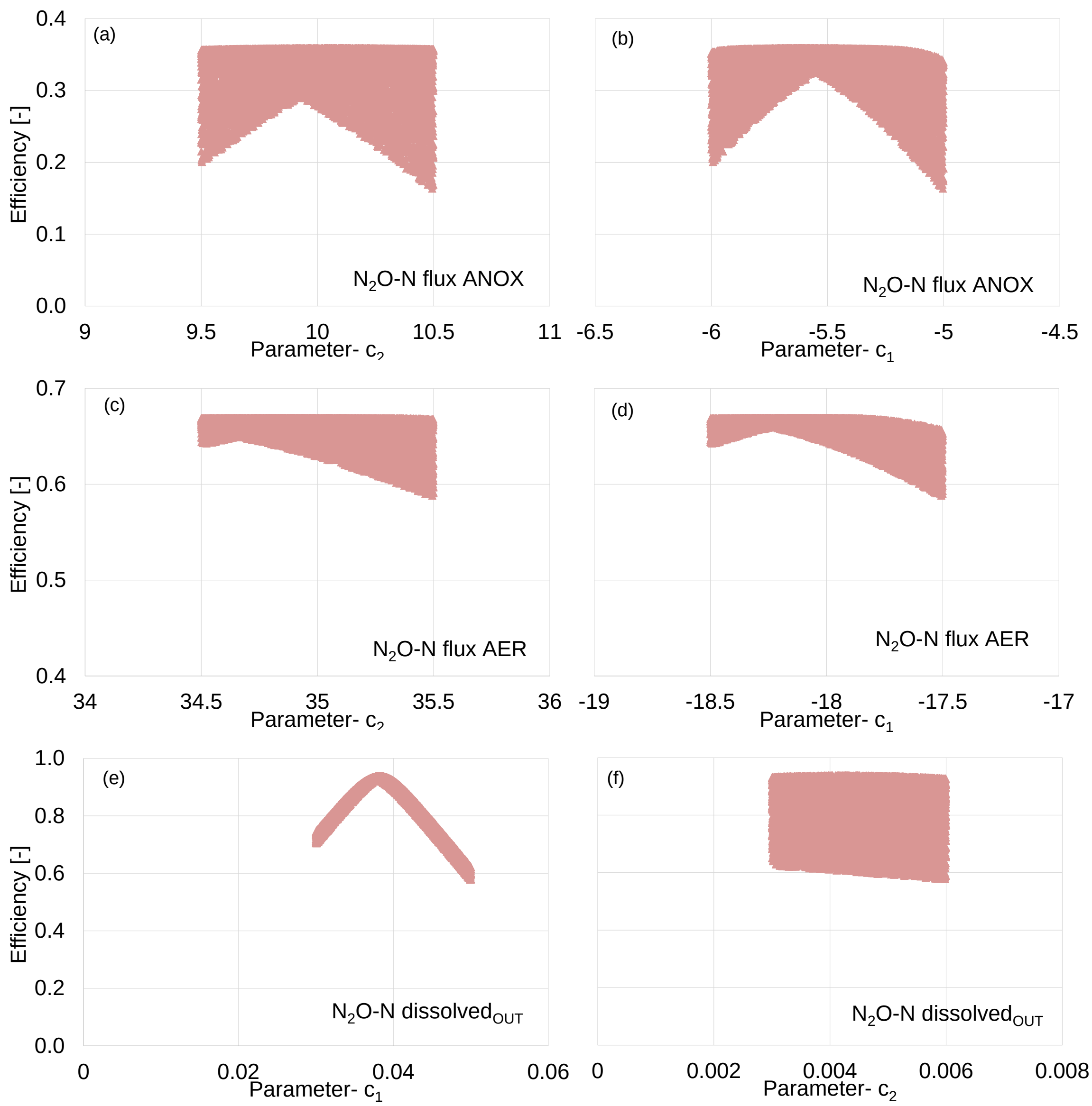

Figure 2. Scatterplot of N_2O-N $flux_{ANOX}$ (a, b) , N_2O-N $flux_{AER}$ (c, d) and N_2O-N $dissolved_{OUT}$ (e, f) for the Phase III.

Such results suggest that in case of indefinite SRT the accumulation of nitrite has to be controlled. Indeed, high NO_2 inside the aerobic tank can favour the AOB denitrification with the consequence production of N_2O (among others, Colliver and Stephenson, 2000). On the other hand, the NO_2 accumulation in the anoxic tank can lead to a decrease of the denitrification rate with the consequent accumulation of NO and N_2O (Kampschreur et al., 2009). A similar trend for the N_2O-N flux from the anoxic tank was obtained for the Phase II (SRT = 30 days) with an efficiency of 0.6 (Table 2). It is worth to note that during the Phase II (SRT = 30 days) the N_2O-N $dissolved_{OUT}$ mainly depends on the COD_{OUT} (with an efficiency of 0.72, Table 2). More precisely, with the increasing of the COD_{OUT} the N_2O-N $dissolved_{OUT}$ value increases reduced of 0.109 which represents the value of c_2 (Table 2). Such a result suggests that the dissolved N_2O in the permeate (N_2O-N $dissolved_{OUT}$) can be simply predicted by measuring the effluent COD or can be reduced improving the processes aimed at the carbon biological removal (which include denitrification). Finally, by analyzing data of Table 2 one can also observe that the highest absolute efficiency (as the sum of the efficiency of all the independent variables investigated) was obtained during the Phase III (SRT = 15 days). This is mainly related to the fact that during the Phase III a lower variability of the measured data occurred. The lowest SRT value makes the conditions of N_2O production more sharped than the other two phases. Indeed, a clear dependence of N_2O-N dissolved$_{OUT}$ on the NO_2-N_{AER} with an efficiency of 0.94 can be observed during the Phase III. Such a dependence is mainly due to the fact that at low SRT the nitrite accumulation during nitrification takes place (Van Loosdrecht and Salem, 2006). For sake of completeness, in Figure 2 the scatter plots are reported which show the efficiency value for each Monte Carlo simulation of some dependent variables for the Phase III. By observing the scatter plots of Figure 2 the clear dependence of N_2O-N dissolved$_{OUT}$ on the NO_2-N_{AER} can be also seen (Figure 2e). Further, the variation of the parameter c_2 seems to poorly influence the efficiency of N_2O-N dissolved$_{OUT}$ thus suggesting that c_2 should also be null to predict the N_2O-N in the permeate. For the other dependent variables the scatter plots of Figure 2 show a sort of combining effect due to the variation of both c_1 and c_2 on the dependent variable efficiency.

Complex multiregression analysis

On the basis of the knowledge acquired by means of the simple linear regression analysis, the complex mutiregression analysis has been performed by considering all the data acquired during the three experimental phases. Therefore, the analysis has been performed taking into account the specific operational conditions of each phase. Only two dependent variables have been considered for the complex mutiregression analysis: the sum of the N_2O-N flux emitted from each tank ($\sum N_2O$-N flux) and the N_2O-N permeate dissolved concentration (N_2O-N dissolved$_{OUT}$).

Table 3 and Table 4 report the results of the complex mutiregression analysis for each investigated equation related to the obtained maximum efficiency. Data reported in Table 3 show that the LINm equation poorly reproduces the measured data having a low maximum efficiency value (namely, 0.015) without considering the dependency on SRT, pH_{AER} and pH_{ANOX} (Table 3). Conversely, the efficiency obtained with the LINm equation for the N_2O-N dissolved$_{OUT}$ is slightly higher than for $\sum N_2O$-N flux (equal to 0.244) (Table 3). As reported in Table 4, overall poor efficiency values were obtained by applying also the other two equations (EXP and SumEXP) for both the investigated dependent variables. The poor results suggest that an unique expression valid for different operational conditions cannot likely be established due to the interactions among the key factors affecting the N_2O production/emission which differ with the operational conditions (and in particular with the SRT). Indeed, as demonstrated during the simple linear regression analysis the dependence of each N_2O variable varies with the operational conditions and with the key processes occurring inside the tank taken into account.

Table 3. Efficiency and regression coefficient values obtained by applying the LINm equation for each of the investigated dependent variables

		LINm	
		$\sum N_2O$-N flux	N_2O-N dissolved$_{OUT}$
		Efficiency	Efficiency
Independent variable	Coefficient	0.015	0.244
C/N	c_1	0.1912	0.6912
N-$NH_{4,IN}$	c_2	0.1765	0.0765
TSS	c_3	1.3618	1.3618
Biofilm	c_4	0.1736	0.0013
SRT	c_5	-	2.9785
DO_{AER}	c_6	0.1475	0.0002
N-NO_{2_AER}	c_7	1.1548	0.0014
pH_{AER}	c_8	-	0.0205
DO_{ANOX}	c_9	0.0544	0.0738
N-NO_{2_ANOX}	c_{10}	0.1449	0.0037
pH_{ANOX}	c_{11}	-	-

Table 4. Efficiency and regression coefficient values obtained by applying the EXP and the SumEXP equations for each of the investigated dependent variables

		EXP		SumEXP	
		∑N_2O-N flux	N_2O-N dissolved$_{OUT}$	∑N_2O-N flux	N_2O-N dissolved$_{OUT}$
Independent variable	Coefficient	Efficiency 0.125	Efficiency 0.164	Efficiency 0.198	Efficiency 0.178
C/N	c_1	-0.04614	1.00000	-0.04603	0.10000
C/N	c_2	0.23991	0.10000	0.24013	0.10000
N-$NH_{4,IN}$	c_3	0.00813	0.21200	0.00490	0.00940
N-$NH_{4,IN}$	c_4	0.36912	-0.00014	0.15330	0.10000
TSS	c_5	-	0.01000	-	0.10000
TSS	c_6	-	0.11600	-	0.00000
Biofilm	c_7	0.00128	0.01123	0.00567	1.00000
Biofilm	c_8	0.00993	-0.00010	0.01987	0.10163
SRT	c_9	-	-	-	0.19800
SRT	c_{10}	-	-	-	0.30200
DO_{AER}	c_{11}	0.00239	-	0.00489	0.17207
DO_{AER}	c_{12}	0.00193	-	0.00313	-
N-NO_{2_AER}	c_{13}	0.00018	-	0.00017	-
N-NO_{2_AER}	c_{14}	0.00028	-	0.00030	-
pH_{AER}	c_{15}	-	-	-	-
pH_{AER}	c_{16}	-	-	-	-
DO_{ANOX}	c_{17}	0.00152	-	0.00037	-
DO_{ANOX}	c_{18}	0.00324	-	0.00361	-
N-NO_{2_ANOX}	c_{19}	0.00032	-	-	-
N-NO_{2_ANOX}	c_{20}	0.00004	-	-	-
pH_{ANOX}	c_{21}	-	-	-	-
pH_{ANOX}	c_{22}	-	-	-	-

CONCLUSIONS

This paper reports the results of a multiregression analysis performed by adopting data acquired in a UCT-MB-MBR pilot plant. The pilot plant was operated according to three different SRT (namely, ∞, 30 and 15 days, respectively). The multiregression analysis has been performed by adopting both simple (namely, linear) and complex equations. The study was proposed as the first attempt to provide a simple and easy to be used tool for predicting the N_2O emitted from WWTP.

Reasonable agreements were obtained comparing simulated and measured data when simple linear regression equations were adopted. Indeed, the results revealed that the dependency of the N_2O flux changes with the SRT and with the section of the pilot plant under study. The phase at the lowest SRT (Phase III) has provided the highest absolute efficiency. On the other hand, regarding the complex multivariate regressions, results revealed that none of the investigated equations is able to reproduce the measured data with a satisfactory efficiency. Such a result suggested that most likely an unique equation valid for different operational conditions cannot be established. Indeed, the interactions among the key factors affecting the N_2O production/emission differ with the operational conditions (and in particular with the SRT).

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