An integrated sensor for the monitoring of aerobic and anoxic activated sludge activities in biological nitrogen removal plants

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Abstract An integrated sensor is developed as a tool for monitoring the activated sludge activity on which the performance of the treatment plant depends. The sensor provides information-rich data of high frequency obtained from respirometric-titrimetric and nitrate measurements in one single set-up. The sensor is shown to successfully monitor and provide in-depth insight into nitrification, denitrification and carbon source degradation processes occurring in BNR plants. Based on the experimental results it is hypothesized that the ratio of NUR to OUR rather reflects the rate of carbon source uptake (storage) under anoxic and aerobic conditions than growth process.

Keywords Activated sludge; monitoring; nitrate uptake; nitrate production; sensors; titrimetry

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Introduction

Nitrogen removal is mostly accomplished through biological processes. The most commonly designed process configuration is the combination of nitrification and denitrification processes in aerobic and anoxic environments. For biological treatment, although cheaper than its counterpart (the chemical treatment), the process stability is still a major concern. Coupled with the recent demand due to ever more stringent effluent criteria set by EU guidelines, process stability and performance of the biological treatment plants has been challenged seriously. Modelling and control of the biological processes seems promising to meet these challenges for biological nitrogen removal (BNR) plants. The availability of sensors able to provide online and reliable information is central to the development and further application of process control.

In this context, a number of methods and (bio)sensors have been and still being developed. Respirometry has been shown to be a valuable technique in probing information from the activated sludge processes: e.g. monitoring activated sludge activity (Gernaey *et al*., 2001), determination of inhibition kinetics (Kong *et al*., 1996), characterization of influent wastewater and estimation of biokinetic parameters of the activated sludge process (Kappeler and Gujer, 1992; Vanrolleghem and Spanjers, 1998; Vanrolleghem *et al*., 1999; Petersen *et al*., 2002b). Regarding the anoxic biomass activity, nitrate measurements replace the respirometric methods (McClintock *et al*., 1988; Kristensen *et al*., 1992; Naidoo *et al*., 1998) where nitrate is generally measured offline using traditional colorimetric methods or an autoanalyser at a low measurement frequency (around 5–10 min). In addition, titrimetry – an indirect measurement of pH effect of the biomass on the medium – has been well applied for monitoring and quantifying both anoxic and aerobic activated sludge activities (Ramadori *et al*., 1980; Massone *et al*., 1996; Bogaert *et al*., 1997; Gernaey *et al*., 2002).

In this study, a method based on integrated aerobic and anoxic sensors is developed to monitor the entire activated sludge activity occurring in biological nitrogen removal (BNR) plants. The integrated sensor works by sequentially monitoring the aerobic and anoxic

Table 1 The information matrix delivered by the integrated sensor

Processes	Aerobic heterotrophs	Anoxic heterotrophs	Aerobic autotrophs
Aerobic COD oxidation	OUR and HP		
Denitrification		NU and HP	
Nitrification			OUR, NP and HP

activity of the biomass. The aerobic part of the sensor is based on respirometric–titrimetric measurements (Gernaey *et al.*, 2001) while the anoxic part of the sensor is based on nitrate–titrimetric measurements (Foxon *et al.*, 2002; Petersen *et al.*, 2002a). For nitrate measurements, an ion-selective nitrate electrode is employed to provide online, fast and continuous data. The well known electrode drift phenomenon of the nitrate probe (Pedersen *et al.*, 1990) is overcome by implementing the following automatic in situ calibration procedure: the initial nitrate concentration that should be added to start an anoxic experiment is dosed to the reactor in two steps: in the first step 2/3rd of the nitrate is added and this is followed by the addition of the remaining (1/3rd) nitrate. In this way, two points of nitrate versus electrode potential are available for the in situ calibration.

By merging the aerobic and anoxic sensors in one single set-up, the integrated sensor provides the information-rich matrix of data presented in Table 1. The sensor is tested for three distinctive activated sludge activities: carbon degradation, nitrification and denitrification processes. The quality of the experimental data is analysed and the titrimetric data is interpreted based on the conceptual proton production model of Gernaey *et al.* (2002) and Petersen *et al.* (2002a).

Materials and methods

The set-up of the integrated sensor shown in Figure 1 consists of an aeration (2.5 l) and a respiration (1 l) vessel which is made strictly airtight (Gernaey *et al.*, 2001). A cooling system (Lauda Ecoline E303) is used to control the temperature of the reactors.

Data acquisition, pH control and data processing are implemented by Labview software (National Instruments, NIDAQ 6.9 with AT-MIO-16XE-50 DAQ card and Labview 6.i). The dissolved oxygen is measured by Inpro 6100/120/T/N (Mettler Toledo) type oxygen electrodes, which are connected to Knick Process 73 and Knick Stratos 2401 oxygen transmitters respectively. The pH is measured in the aeration vessel with a HA405-DXK-S8/120 type pH electrode (Mettler Toledo), which is connected to a Knick Stratos 2401 pH transmitter, and nitrate is measured with a type S7/120 ion-selective electrode (Mettler Toledo) in combination with a reference electrode, which are both connected to a Knick Process 73 pH transmitter.

The data acquisition frequency of the sensor is set to 3 seconds. High frequency noise known to be present in the weak analog signals of the electrodes of the set-up are filtered using a low pass Savitzky-Golay least square polynomial filter (Press *et al.*, 1992), through a Labview Matlab script node (Matlab R12, The MathWorks Inc.). The pH is controlled within a narrow pH set-band ± 0.03 as described in detail in Gernaey *et al.* (2001).

Activated sludge monitoring methodology

For the experimental work activated sludge was sampled from a lab scale 80 l SBR reactor (Govoreanu *et al.*, 2002). During the experiments small substrate pulses (e.g. 15–20 ml) of acetate or dextrose (10 g COD/l), ammonium (1 g N/l) and nitrate (10 g N/l) stock solutions were dosed to the activated sludge. All experiments were performed at 15.3 ± 0.1°C. The following experimental procedure is applied for on-line data collection: (1) *initialisation phase:* a sludge sample (3 L) taken from the second aeration phase of the SBR is pumped

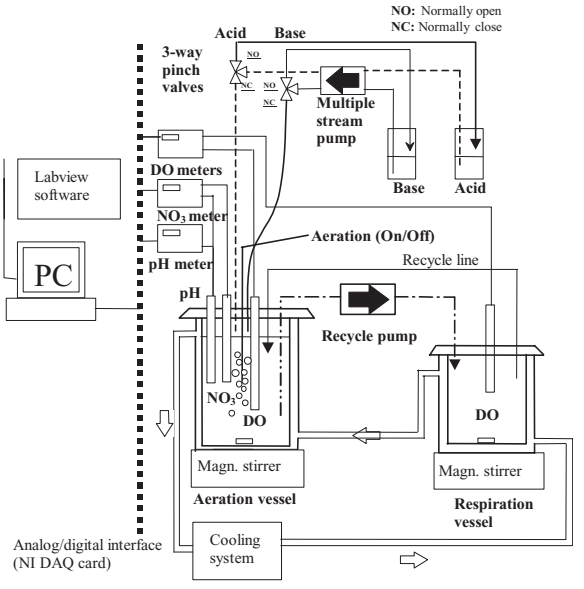

Figure 1 Illustration of the integrated sensor set-up

into the reactor. The sample is brought into the endogenous state; (2) *aerobic experimental phase:* the aerobic cycle is started by addition of a substrate sample (COD, NH_4-N). The pH is fixed to a desired level similar to the operational conditions of the plant. The duration of the cycle is 2–4 h depending on the S_0/X_0 ratio; (3) *anoxic experimental phase:* after the aerobic cycle reaches a steady-state level (relatively constant DO), aeration is switched off. The initial nitrate concentration is measured with the nitrate probe. Nitrate is added step-wise for in situ calibration. This is followed by addition of a COD source (e.g. acetate, dextrose) according to the desired C/N ratio (advised to be sufficiently high, e.g. 8). The duration of the experiment is 2–3 h depending on the C/N ratio; and (4) *discharge of the sample*.

Results and discussion

The potential use of the integrated sensor for monitoring carbon source degradation, nitrification and denitrification processes was evaluated following the experimental procedure outlined above.

Monitoring carbon source degradation processes under aerobic and anoxic conditions

During the aerobic phase, the sensor essentially works according to the methodology of respirometric-titrimetric measurements developed and tested for activated sludge by Gernaey *et al*. (2001, 2002). In Figure 2A and Figure 2B, a typical response of activated sludge to a pulse addition of acetate and dextrose is presented respectively. As expected, the addition of the readily degradable substrate acetate induces a rapid depletion of oxygen in the reactor, which is accompanied by an increased rate of acid addition to keep the pH constant. As explained in detail in the titrimetric model of Gernaey *et al*. (2002), 4 processes basically influence the proton concentration during the course of aerobic acetate degradation: the uptake of carbon source (for weak acids) leads to proton consumption, uptake of ammonia for growth will release protons, CO_2 production as a result of carbon source oxidation will release protons and finally CO_2 stripping from the reactor due to aeration will consume protons from the medium. From the proton production model, it is apparent that two processes dominate the proton consumption from the medium (which is compensated

by acid addition to keep the pH constant): uptake of acetate and CO_2 stripping from the medium. In Figure 2A, two acid addition rates can be discerned: one corresponding to rapid uptake of acetate by biomass plus CO_2 stripping and the second one is the acid addition rate due to the CO_2 stripping effect. In other words, the increased rate of cumulative acid addition indicates an increased rate of acetate uptake, which ceases once all the acetate is consumed. From that point on, acid addition is required mainly to compensate for the proton consumption by the CO_2 stripping process, which is assumed to be constant for short-term experiments (Gernaey *et al.*, 2002). It is known, however, that the acid addition rate due to CO_2 stripping will decrease exponentially in the long term as the bicarbonate system in the medium tends to attain equilibrium (Iversen *et al.*, 1994).

The oxygen uptake rate of the biomass attains its maximum in a very short time after the addition of the acetate. The observed transient delay (2 to 5 minutes in Figure 2A) in reaching the maximum activity is typical for batch experiments and is conceived to be the start-up phenomena of the biomass as discussed by Vanrolleghem *et al.* (1998).

The activated sludge response to dextrose addition presented in Figure 2B demonstrates the effect of the carbon source on the titrimetric profile. Contrary to the acetate addition, the uptake of dextrose (present in bulk liquid in undissociated form) induces no effect on the proton concentration in the medium. In this case, the ammonia uptake for growth and CO_2 production processes release protons to the medium, which is accompanied by a decreased rate of acid addition during the dextrose uptake phase. After all the dextrose is consumed from the medium, the acid addition increases immediately to compensate for the proton consumption due to CO_2 stripping (Gernaey *et al.*, 2002).

The response of activated sludge to pulse additions of acetate and dextrose under anoxic conditions is presented in Figure 2C and Figure 2D respectively. Under anoxic conditions, the denitrifying fraction of the biomass starts immediately to take up acetate (shown by acid

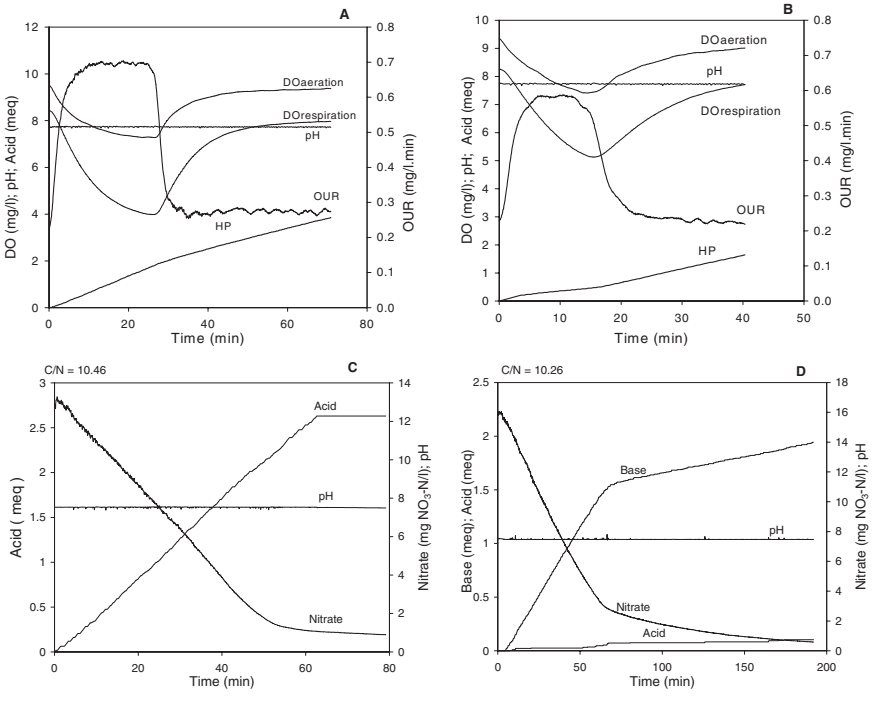

Figure 2 Typical experimental results from the integrated sensor. The aerobic phase of the sensor: 15 ml acetate addition with pH set point 7.7 (A), 15 ml dextrose addition with pH set point 7.5 (B). The anoxic phase of the sensor: acetate addition with C/N 10.46 and pH set point 7.7 (C), dextrose addition with C/N 10.26 and pH set point 7.5 (D)

addition in Figure 2C) and use nitrate. Since CO_2 stripping is quite small (Bogaert *et al*., 1997) during anoxic experiments but still cannot be assumed to be zero due to surface mass transfer, the dominant effect on the proton consumption is induced by acetate uptake. In addition to acetate uptake, uptake of nitrate and nitrite as electron acceptors will induce proton consumption from the medium too as explained in detail in Petersen *et al*. (2002a). The acid addition ceases when all the acetate is taken up from the medium (Figure 2C), while nitrate reduction continues, albeit at a drastically lowered rate.

The anoxic response of activated sludge to dextrose addition is a base addition (Figure 2D), in contrast to the acetate, which agrees with the proton production model of Petersen *et al*. (2002a). Apparently, in this case, the proton production processes, which are ammonia uptake and CO_2 production, dominate the proton concentration in the medium. The exogenous nitrate reduction (i.e. the nitrate reduction by exogenous carbon source) is fast and follows zero order kinetics (indicating the Monod terms for substrate and other growth limiting nutrients are close to unity). After the external carbon source (dextrose) is completely removed from the mixed liquor, as indicated by the breakpoint in the base addition profile and the nitrate reduction curve, the base addition as well as the nitrate reduction rates continue at a lower rate for quite a long time (over 100 min). The explanation for these phenomena could be either endogenous nitrate reduction or nitrate reduction due to endogenous processes and degradation of internally stored products (Daigger and Grady, 1982). The latter seems to be plausible when considering the history of the sludge. The SBR is operated batch-wise, where the biomass is exposed to feast and famine periods. It is a well observed fact that the response of activated sludge to this environment is to form storage products (Daigger and Grady, 1982; Dircks *et al*., 2001; Dionisi *et al*., 2001).

The experimental results obtained at lower C/N ratios (presented in Figure 3) support this hypothesis. As shown in Figure 3 (right), base addition accompanies the nitrate reduction by dextrose, which ceases once the external dextrose is removed from the medium. The base addition after a pause period (a number of minutes depending on the bandwidth of the pH controller) swaps to acid addition, indicating a change in the dominance of the pH affecting processes. Preliminary simulations with the conceptual proton production model of Petersen *et al*. (2002a) indicate that the endogenous process causes slight proton consumption from the medium (increase in alkalinity). Based on these arguments, it can be concluded that the activated sludge seems to reduce nitrate (and other denitrification intermediates) by the internally stored products at a quite lower rate than the exogenous nitrate reduction rate. The anoxic phase of the integrated sensor is thus able to successfully monitor the response of the activated sludge under anoxic conditions for various substrate sources including storage phenomena.

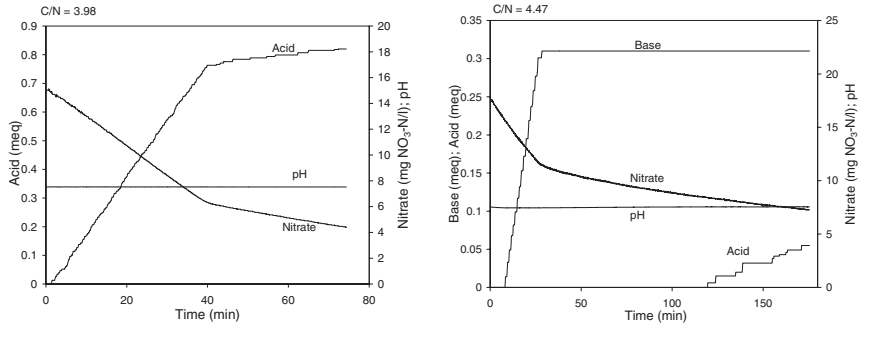

Figure 3 Anoxic activated sludge response to low COD/N conditions: acetate addition with C/N 3.98 and pH set point 7.5 (left), dextrose addition with C/N 4.47 and pH set point 7.5 (right)

Table 2 Anoxic activated sludge monitoring results for exogenous and endogenous processes

	Exogenous		Endogenous	
	NUR (mg NO_3-N/l-min)	HPR (meq H^+/min)	NUR (mg NO_3-N/l-min)	HPR (meq H^+/min)
Acetate (high C/N)	0.2347	–0.0439	–	–
Acetate (low C/N)	0.22578	–0.01978	0.05356	–0.0016
Dextrose (high C/N)	0.21935	0.02537	0.02939	0.0034
Dextrose (low C/N)	0.2275	0.01229	0.0261	0

Anoxic reduction factor of heterotrophic growth. The anoxic growth reduction factor (Henze *et al.*, 1987) becomes identifiable straightforwardly with the online information obtained from the integrated sensor by comparing the aerobic (OUR) and anoxic heterotrophic activities (NUR) as follows: $\eta_g = 2.86 \times \text{NUR/OUR}$, where NUR is the nitrate uptake rate of the biomass, determined from the derivative (slope) of the nitrate uptake curve. The η_g for acetate and dextrose is found to be 0.953 and 1.06 respectively. The results suggest that the biomass is very well able to denitrify under anoxic conditions.

The nitrate uptake rate of biomass (NUR) given in Table 2 is observed not to change under high and low C/N ratios, which seems reasonable as the major fraction of the biomass is expected to favour carbon source uptake under aerobic and anoxic conditions rather than growth (Daigger and Grady, 1982; van Loosdrecht and Heijnen, 2002). In fact, considering the anoxic reduction factor (the ratio of NUR to OUR) is approximately 1 implies that the growth is not affected under anoxic conditions. However, model based interpretation of aerobic and anoxic data (results not shown) showed that the yield coefficient under aerobic and anoxic conditions is considerably higher than the accepted default value of 0.67 for heterotrophs (Henze *et al.*, 1987). Under these conditions, the comparison of NUR and OUR will not truly reflect the anoxic growth reduction. Rather, it is expected to determine the ratio of NUR to OUR dominated by the storage rate under anoxic (Dionisi *et al.*, 2001) and aerobic conditions (Dircks *et al.*, 2001). Therefore, it may seem reasonable to conclude that the uptake rate of carbon source remains equal in aerobic and anoxic conditions. It is also observed that the NUR after the external carbon source is removed from the medium is different for experiments with acetate and dextrose. This suggests that the utilisation rate of stored products with dextrose is slower than that of acetate. This is in accordance with the findings of Dircks *et al.* (2001). It is clear that these arguments need to be confirmed with dedicated experiments.

However, it is not clear as to why the proton production rate determined from the slope of the cumulative acid (acid addition means proton consumption) or base addition (proton production) curves (HPR) is halved for low C/N ratios. This point is currently being investigated with a mathematical model developed to simulate the experimental results.

Monitoring simultaneously nitrification and carbon source degradation kinetics

Combining the nitrate electrode with the respirometric-titrimetric measurements offers the opportunity to monitor the nitrification and carbon degradation processes in a single experiment as presented in Figure 4, in this way excluding the need to perform at least two independent experiments (one with nitrification inhibitor (e.g. ATU) addition and the other without inhibiting the nitrifiers (Kong *et al.*, 1996)). Cross-analysing the OUR, titrimetric data and nitrate shows that the acetate uptake and the nitrification start immediately and reach their maximum conversion rate after a start-up phenomenon. The additional availability of nitrate measurements provides an independent observer that can be used to at least qualitatively check the results of simultaneous carbon source degradation and nitrification kinetics.

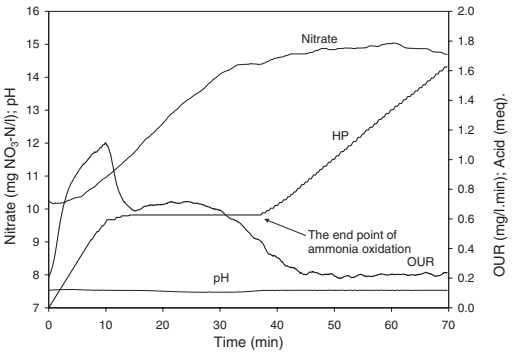

Figure 4 Monitoring carbon source degradation and nitrification processes in single batch reactor: pulse addition of 8.5 ml acetate and 15 ml ammonium nitrogen with pH-set point 7.5

The titrimetric profile obtained from this experiment confirms the proton production model of Gernaey *et al*. (2001, 2002). During the first phase, increased acid addition occurs due to rapid acetate uptake. In the second stage, no acid and base addition is necessary due to the equilibrium between two competing pH-affecting processes: ammonium oxidation (first step nitrification) releases protons that are compensated by the proton consumption due to CO_2 stripping. Soon after the endpoint of the 1st step nitrification, the acid addition is resumed due to the CO_2 stripping effect.

Conclusions and perspectives

The integrated sensor developed here essentially is a tool for monitoring activated sludge activities occurring in BNR plants in a single set-up. The integrated sensor is shown to successfully monitor nitrification, denitrification and aerobic carbon source degradation processes. The information rich matrix of data (OUR, HP and NU or NP) provided by the integrated sensor can be used to quantify and estimate activated sludge activities. Furthermore, each independent measured variable can be used as an internal check for the quality of the experimental results. The information provided by the sensor (titrimetry mainly monitors the carbon source uptake while OUR and NU monitor the energy generation of the biomass) is shown to be equally powerful in studying the quite significant storage response of the activated sludge in batch experiments for anoxic and aerobic conditions. The conceptual proton production model proposed elsewhere was shown to explain the titrimetric results well. Based on the experimental results it is hypothesized that the ratio of NUR to OUR rather reflects the rate of carbon source uptake (storage) under anoxic and aerobic conditions than the energy generation process. This reduction factor was close to 1 for the activated sludge under study, which means that carbon source uptake was similar in both conditions.

Acknowledgement

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Nomenclature

C/N	Initial COD to nitrogen ratio (g COD/g N)
COD	Chemical oxygen demand
HP	Cumulative proton production (meqH^+)
HPR	Proton production rate (meq H^+/min)
NU	Nitrate uptake (mg NO_3-N/l)

NP	Nitrate production (mg NO_3-N/l)
NUR	Nitrate uptake rate (mg NO_3-N/l-min)
NH_4^+-N	Ammonium (concentration) (mg N/l)
S_0/X_0	Initial substrate to biomass ratio (gCOD/gCOD)
OUR	Oxygen uptake rate (mg O_2/l-min)
η_g	Anoxic growth reduction factor

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