New urban wastewater treatment with autotrophic membrane bioreactor at low chemical oxygen demand/N substrate ratio

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ABSTRACT

The potential for total nitrogen removal from municipal wastewater has been evaluated in an autotrophic membrane bioreactor running with a low chemical oxygen demand (COD)/N ratio to simulate its combination with an upstream physicochemical process that retains a large proportion of organic matter. The tests were conducted in a laboratory scale submerged membrane bioreactor loaded with a synthetic influent. Nitrogen loading rate was 0.16 $kg_{N-NH4+}.m^{-3}.d^{-1}$ and sodium acetate was added as a carbon source. Results have shown that nitrogen elimination can reach 85% for a COD/N ratio of 5, with COD removal exceeding 97%. However, a COD/N ratio of 3.5 was found to be the limiting factor for successfully reaching the overall target value of 10 $mgN.L^{-1}$ in the effluent. Nevertheless, low COD/N ratios make it possible to work with low total suspended solid concentrations in the bioreactor, which greatly facilitates membrane fouling control by a simple aeration and backwashing strategy.

Key words | activated sludge population dynamics, autotrophic membrane bioreactor, COD/N ratio, energy demand, nitrogen removal

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INTRODUCTION

There is an increasing need to use reliable, efficient and sustainable technologies to (i) treat wastewater and obtain a sufficient quality of treated water for reuse, (ii) recover molecules of interest and eventually (iii) produce sustainable energy. Urban wastewater treatment plants are defined today to remove particulate and dissolved organic fractions and, in more sensitive areas, nitrogen and phosphorus compounds. The most conventional well-known intensive system to treat urban wastewater is the activated sludge process.

Recent studies have shown the potential of intensification (in terms of treated water quality and unit compactness) by replacing the settling separation phase with a selective membrane barrier, which gives rise to a membrane bioreactor (MBR) (Grasmick *et al.* 2007). Nevertheless the main but significant problem with MBR is its major energy requirements due to (i) aeration for aerobic reactions in concentrated biological suspensions and (ii) aeration to control membrane fouling (Krause & Cornel 2007; Lebègue *et al.* 2009; Kraume & Drews 2010).

A new concept is thus proposed for an MBR presence in an intensive wastewater treatment plant that will show a global positive energy balance. It is based: (i) 'in the water line', associated with a combination of an efficient upstream liquid/solid separation and coagulation/flocculation with a laminar settler or a drum filter, for example, able to remove and concentrate organic matter in the primary sludge (An *et al.* 2009), and an autotrophic membrane bioreactor (AutoMBR) able to remove nitrogen, residual soluble organic matter and germs; and (ii) 'in the sludge line', on an anaerobic digester able to recover methane and provide energy from fresh primary sludge. The AutoMBR then receives an influent containing mainly the residual soluble fraction of organic carbon present in domestic wastewater and a large part of the nitrogen fraction that escapes from the primary treatment. Therefore, the AutoMBR influent presents a low chemical oxygen demand (COD)/N ratio (depending on the performance of the upstream primary treatment) favorable to the nitrification stage (as nitrification is carried out by autotrophic bacteria,

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Li *et al.* 2006) but which may be an obstacle for sufficient denitrification (Deronzier *et al.* 2001; Sun *et al.* 2010).

The residual organic matter is only used for denitrification when the COD/N ratio is sufficiently low and oxygen is only necessary for nitrification. The intensity of aeration in the bioreactor then appears significantly lower (ASCE 1992). Moreover, air requirements to minimize membrane fouling could be lower due to lower biomass concentration and activity in the bioreactor bulk phase. At last, the global system can then show a positive energy balance because fresh settled primary sludge is able to generate more biogas (and energy) than secondary sludge coming from a conventional system. It is one of the challenges of this research.

This paper is mainly focused on four points: (i) identification of the critical COD/N ratio that allows sufficient nitrogen removal by nitrification/denitrification; (ii) the biological kinetics and treatment performance; (iii) the oxygen requirements; and (iv) the potential gain in the suspension's filterability by reducing the growth of heterotrophic populations, due to the low concentration of organic carbon in the AutoMBR influent (Mengchun *et al.* 2004).

MATERIAL AND METHODS

Membrane bioreactor operation

The experimental laboratory scale bioreactor was composed of two tanks with identical volumes of 30 L connected in series. One worked in anoxic conditions; it received the influent containing soluble organic matter and ammonium as the nitrogen source. The second tank was aerated and equipped with submerged flat sheet membranes with a pore size of 0.04 μm and a surface area of 0.34 m^2 (Microdyn-Nadir, Germany). Air diffusers were set below the membrane module; the aeration not only supplied oxygen to microorganisms, but also produced hydraulic shearing forces close to the membrane surface to delay membrane fouling. The treated water was recovered by filtering the biological suspension present in the aerated tank through the membrane barrier. The trans-membrane pressure (TMP in kPa) was measured by specific pressure sensors placed upstream and downstream of the membranes. Nitrates were formed in the aerated tank by ammonium oxidation. To ensure nitrate reduction, the mixed liquor present in the aerated tank was recycled towards the first anoxic tank. The recycle ratio (R) was set at 400% relative to the influent flow.

The autotrophic submerged membrane bioreactor (AutoMBR) was fed with a synthetic substrate containing 70 $mg_{N\text{-}NH4+}.L^{-1}$ diluted in tap water. This value is close to the R1 nitrogen concentration measured in raw domestic wastewater. Acetate at different concentrations (from 245 to 560 $mg_{COD}.L^{-1}$) was added to the AutoMBR influent to simulate the presence of soluble organic matter coming from the primary treatment. According to the performance of the primary treatment, the COD/N ratio influence was studied for successive values, mainly 5 and 3.5 (in place of the COD/N ratio from 10 to 12 encountered in domestic wastewater). The bioreactor was inoculated with seed sludge taken from an activated sludge process treating domestic wastewater under extended aeration (sludge age close to 20 days). Experiments were performed during a period of 180 days with various solids retention times (SRT), hydraulic retention times (HRT) and influent concentrations, as detailed in Table 1.

Table 1 | Operating parameters of the AutoMBR during the 180 days of operation

	Phase 1 D0–D15 starting phase	Phase 2 D16–D22 MBR	Phase 3 D23–D75 MBR	Phase 4 D76–D85 batch	Phase 5 D86–D105 reduction of flow	Phase 6 D106–D108 recovery of flow	Phase 7 D109–D180 MBR
SRT (d)	No extraction	20	**40**	No extraction	40	40	**40**
HRT (h)	12.6	12.6	**12.6**	No filtration	25.2	25.2–12.6	**12.6**
Filtration flux J_W ($L.h^{-1}.m^{-2}$)	14	14	**14**	–	7	7 → 14	**14**
Organic loading rate ($kgCOD.m^{-3}.d^{-1}$)	1.27	0.81	**0.81**	0	0.28	0.56	**0.56**
Nitrogen loading rate ($kgN. m^{-3}.d^{-1}$)	0.16	0.16	**0.16**	0.03	0.08	0.16	**0.16**
COD/N	8	5	**5**	0	3.5	3.5	**3.5**

Phases 1 and 2 corresponded to biomass acclimatization to the new operating conditions. Phases 4–6 were developed to analyze specific aspects of the process not described in this document. The results discussed in this paper correspond to data obtained during phases 3 and 7.

The AutoMBR was operated at room temperature (16–21 °C) except during 8 days in winter with lower temperature (D68–D70 and D135–D138 with temperatures below 15 °C). The pH of the aerobic tank was regulated at 7.5 ± 0.5 thanks to the addition of a sodium hydroxyde solution (2 mole.L^{-1}) by a pH controller and a dosing pump. Membrane fouling was controlled by air diffusion and a specific filtration strategy composed of successive 10-min operational filtration cycles including 8.75 min of suction (flow rate of 17 LMH (L.h^{-1}.m^{-2})) followed by 0.25 min of relaxation (no filtration), 0.75 min of reverse-washing (flow rate of 9 LMH) and 0.25 min of relaxation. Thus the average effective flow rate was close to 14 LMH. A storage tank was present on the effluent line in order to provide a clean water reserve for backwashing phases.

MODELING

Activated sludge model 1 (ASM1) was used to simulate the operations (Henze et al. 2008). ASM1 was used with default kinetic parameters at 20 °C. Both tanks were assimilated to perfectly stirred reactors, and the membrane separation step was considered as using a perfect particle separator with a negligible volume.

Analytical methods

Approximately once a week, sludge samples were taken to analyze the evolution of the biological population. Total genomic DNA extraction from the AutoMBR samples was carried out following the method detailed in Godon et al. (1997). The DNA extracts were purified with the QIAamp DNA Mini Kit (Qiagen). Fingerprints of bacterial communities were generated by capillary electrophoresis-single strand conformation polymorphism (CE-SSCP) (Zumstein et al. 2000). The StatFingerprints library from R (Michelland et al. 2009) was used to analyze CE-SSCP profiles. The fingerprints were first aligned with the internal standard ROX and the sum of the peak areas was normalized to unity. The complexity of the bacterial community was estimated using Simpson's diversity index, by considering the number of species (number of peaks) as well as their relative abundance (area under each peak).

The concentration of mixed liquor suspended solids (MLSS), mixed liquor volatile suspended solids (MLVSS) and COD were measured using standard methods: AFNOR NFT 90–105, NFT 90–029 and NFT 90–101 respectively.

Nitrogen compounds (NH_4^+-N, NO_2^--N and NO_3^--N) were quantified by spectrophotometric analysis (salicylate method by Amver 26069-45 Test N′Tube™ and cadmium reduction method by NitraVer® 5 tests, Hach, Loveland, CO, USA). In addition, specific probes were used to continuously measure pH, temperature, ammonium and nitrate ions in the mixed liquor (Wissenschaftlich-Technische Werkstätten GmbH, Germany). The suspension was also characterized through conventional particle size measuring, soluble microbial products (proteins and polysaccharides in solution) and settleability index.

RESULTS AND DISCUSSION

Suspended solids

Figure 1 shows the evolution of MLSS and MLVSS from September 2012 to March 2013. During phase 2 and the beginning of phase 3 (transient period of phase 3) no sludge extraction was practiced, in order to reach steady-state conditions quickly. During phase 6, the organic loading rate (OLR) was drastically increased, modifying then the growth rate of the biomass until the tenth day of phase 7 (transient phase 7). This time required for stabilization in phase 7 was probably due to the time required for assimilation of excess substrate stored by the biomass during phase 6. After the first transient period in phase 7, a steady state was observed.

Experimental results (MLSS concentrations in the bulk phase) appear in agreement with ASM simulations during steady-state periods of phases 3 and 7 (simulations are represented by the dotted line in Figure 1).

Nitrogen removal

Experimental and modeling results are listed in Table 2 for steady-state periods of phases 3 and 7. During these two phases, the NH_4^+-N concentration in the influent was set at 70 mgN.L^{-1}.

During phase 3 (COD/N = 5), the experimental values of NH_4^+-N and NO_x-N concentrations for anoxic and aerated tanks are in agreement with modeling simulations. During phase 7 (COD/N = 3.5), experimental values of

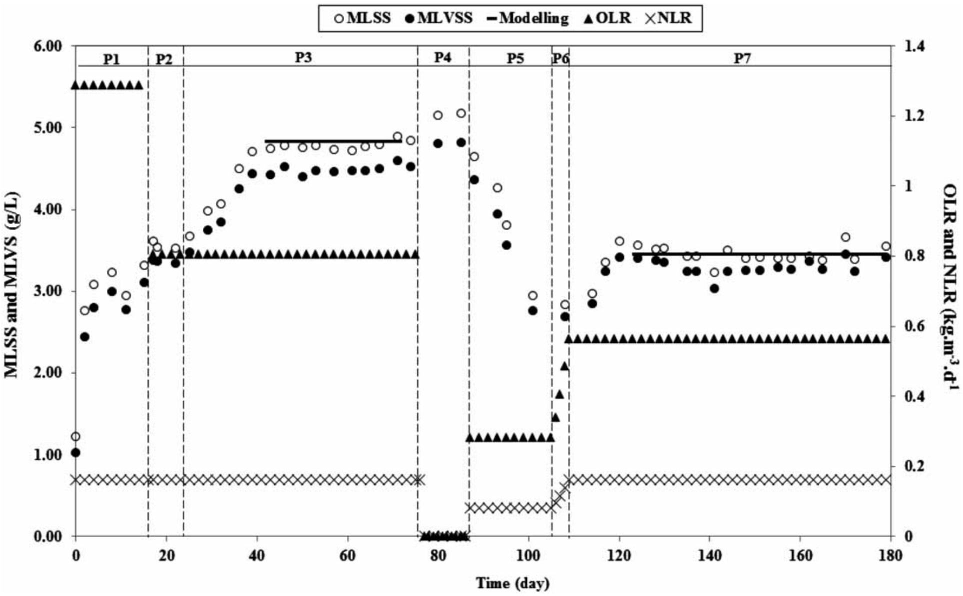

Figure 1 | MLSS and MLVSS concentrations ($g.L^{-1}$) for different organic and nitrogen loading rates: experimental data (symbols) and modeling results (line) during the 180 days of AutoMBR operation.

Table 2 | Values of biological kinetics and conversion rate (experimental data and ASM1 modeling results)

	Phase 3			**Phase 7**		
	Transient	**Permanent**		**Transient**	**Permanent**	
		Experiment	**Modeling**		**Experiment**	**Modeling**
OLR ($kgCOD.m^{-3}.d^{-1}$)	0.81	0.81	0.81	0.56	0.56	0.56
NLR ($kgN.\ m^{-3}.d^{-1}$)	0.16	0.16	0.16	0.16	0.16	0.16
COD/N	5	5	5	3.5	3.5	3.5
biomass growing rate: r_x ($g.L^{-1}.d^{-1}$)	0.122	0.12	0.121	0.146	0.082	0.086
Net growing rate: μ_{app} (d^{-1})	0.03	0.025	0.025	0.043	0.025	0.025
Yield coefficient for biomass growth on substrate: Y_{obs} ($kg_{MLVSS}.kg_{COD}^{-1}$)	0.151	0.148	0.15	0.26	0.145	0.154
$N\text{-}NH_{4effluent}$ ($mgN.L^{-1}$)	21	0.5	0.2	0.6	0.6	0.2
$N\text{-}NO_{3effluent}$ ($mgN.L^{-1}$)	15.5	11.8	12.8	13.3	12.5	27.6
Ammonium oxidation rate ($kgN\text{-}NH_4^+.\ m^{-3}.d^{-1}$)	–	0.39	0.25	–	0.44	0.29
$kgO_{2consumed}.\ m^{-3}$ water	–	<0.32	–	–	0.32	–

NH_4^+-N concentration for the two tanks are slightly greater than values expected by modeling. By contrast, there is a significant difference between experimental results and modeling for NO_x-N, which is due to the fact that ASM1 has simulated an insufficient level of denitrification with such a low COD/N ratio.

Due to a relatively high pH observed in the anoxic tank (pH = 8.5–8.7), a part of the NH_4^+-N was probably transformed into NH_3-N, which is an inhibitor for nitrifying *Nitrobacter* populations (Yoo *et al.* 1999). As a result, the total oxidation of NH_4^+-N to NO_3^--N was not achieved in the aerated tank, leading to the accumulation of NO_2^--N compounds. Such a low COD/N ratio appeared sufficient for the reduction of N-NO_2^- in N_2 in the anoxic tank since the N-NO_3^- shortcut allows a potential saving of 33% of the COD required (Ruiz *et al.* 2006). ASM simulation has

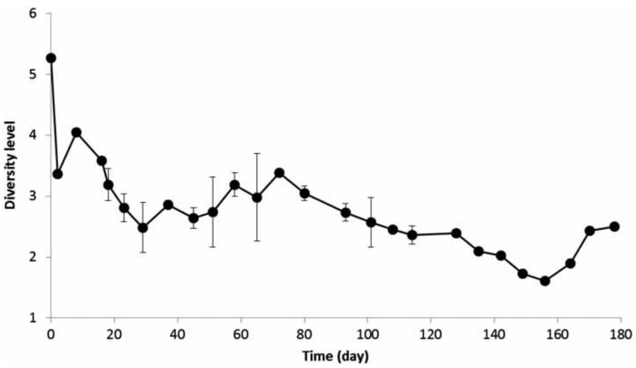

Figure 2 | Temporal dynamics of the diversity of bacterial communities in the AutoMBR as represented by Simpson's diversity index. The error bars represent the standard deviation between measurements in denitrification and nitrification tanks.

not taken into account such insufficient pH control, *Nitrobacter* inhibition or nitrates shortcut, which explains the important differences between the modeling simulation and experimental results.

Kinetic coefficients

The MLSS and MLVSS monitoring allowed the quantification of some apparent kinetic coefficients of apparent cell growth (Table 2).

Except during the transient period at the beginning of phase 7, apparent r_x and biomass concentration increased with OLR (nitrogen loading rate (NLR) was constant) but μ_{app} and Y_{obs} did not really change with OLR because the SRT was maintained at a constant value of 40 days.

The capacity of ammonium oxidation by the active biomass was measured experimentally by punctual impulsions of ammonium in a batch reactor filled with biomass taken from the AutoMBR. It was compared with ASM1 simulation results and NLR imposed on the AutoMBR. As with the ASM1 simulation results, it appeared higher than NLR but this did not indicate that nitrification was not achieved with a low COD/N ratio.

As expected, oxygen requirements including nitrogen oxidation and biomass respiration were significantly lower than in conventional MBR (about twice lower).

Fouling dynamics

The dynamics of membrane fouling were quantified through the evolution of the TMP. During 180 days of operation only two chemical cleanings were performed. Conversely, when the biological process was stabilized (phase 3), the TMP increased slowly and did not exceed 0.15 kPa.d^{-1} which meant a daily variation of total hydraulic resistance close to 0.03 m^{-1}.d^{-1}. The evolution of TMP was in the low range of values observed in the literature except under specific conditions:

- During acclimatization to new conditions (phases 1 and 2) and significant difference between the COD values in supernatant and permeate, the rate of TMP evolution was 0.23 kPa.d^{-1} (eight times higher).
- During abrupt change of filtration conditions (the permeate flux was doubled at the beginning of phase 6), the rate of TMP evolution was 1.62 kPa.d^{-1} (50 times higher).

Bacteria community diversity

The AutoMBR conducted in this study consisted of unusual environmental conditions for bacterial communities mainly because of the long SRT and the low COD/N ratio. Investigating the dynamics of bacterial communities in this original ecological niche is thus necessary for improving our understanding of this innovative process. The temporal dynamics of AutoMBR bacterial communities' diversity is revealed by CE-SSCP fingerprints as presented in Figure 2. Thereafter, during the overall operation of the AutoMBR, the diversity level varied between 1.8 and 4, which is in the range of that found in nitrifying particulate biofilms fed at COD/N = 0 (Gevaudan *et al.* 2012). During the acclimatization period, the diversity level dropped from 5.3 (inoculum) to 4.1 ± 0.9, as an adaptation to new environmental conditions (carbon source, organic load, SRT, HRT). The diversity level then stabilized at 2.9 ± 0.3 during phase 3 (at an SRT of 40 days and a COD/N ratio of 5). Thereafter, the decrease of the COD/N ratio to 3.5 was associated with a decrease in the diversity of the bacterial communities. The Simpson index over the period of operation at a COD/N ratio of 3.5 (phases 5–7) was 2.3 ± 0.4, which is significantly lower from that at a COD/N ratio of 5 (ANOVA (analysis of variance) test, $p = 0.0005$). Our results thus demonstrate that the gradient of environmental conditions used in the MBR, from conventional systems to AutoMBRs operated at long SRT and low COD/N ratio, resulted in a simplification of the bacterial communities. This suggests that AutoMBRs operated at long SRT and low COD/N ratio give rise to unusual communities compared with those of conventional systems.

CONCLUSION

The experimental results have shown the potential to achieve high removal efficiencies of nitrogen compounds even with a low COD/N ratio of 5 $g.g^{-1}$. The AutoMBR unit ensures well-known treatment performance for total suspended solids and COD removal, 100% and more than 95% respectively. For total nitrogen, 85% of removal has been obtained with a COD/N of 5 in agreement with ASM1 modeling. When the COD/N ratio decreased to 3.5, the experimental performance of total nitrogen removal was reduced to 82%. However, this result was higher than the value simulated with ASM1 due to a high pH value in the anoxic tank that induced a partial nitrification in the aerated tank, limiting *Nitrobacter* development. Nevertheless it is important to note the low biomass concentration in AutoMBR when working under a low COD/N ratio and the notable associated kinetic coefficients.

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