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Environmental impact

Municipal wastewater treatment using the membrane bioreactor (MBR) activated sludge process is an increasing environmental practice. However, the treatment efficiency of the MBR process in low-temperature zones is not as high as it is under normal conditions. Understanding the responses of MBR to long-term, continuous low wastewater operation will help guide how MBR wastewater treatment facilities should be operated and managed in cold climate zones to meet stringent discharge regulations. Our work suggests that long-term, low wastewater temperature operation deteriorated the effluent quality but MBR is still a good practice for wastewater treatment in low temperature zones.

Title:

Changes in wastewater treatment performance and activated sludge properties of a membrane bioreactor at low temperature operation

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Abstract

The membrane bioreactor (MBR) activated sludge process is being applied more and more for wastewater treatment due to its high treatment efficiency and low space requirement. However, the usefulness of MBR process in low-temperature zones is less studied than that under normal conditions. This study determined the effect of low temperature (~ 13 °C) operation on MBR performance and activated sludge characteristics. When the wastewater temperature decreased from 22 °C to 13 °C, the average effluent COD concentration increased from (10 ± 5) to (25 ± 4) mg/L and the nitrogen removal efficiency appeared not to be affected. The abundance and diversity of nitrifying bacteria such as *Nitrosospira* (ammonia-oxidizing bacteria) and *Nitrospria* (nitrite-oxidizing bacteria) in the activated sludge were reduced under low temperature exposure. The total biomass concentration decreased from about 10,000 mg COD/L at room temperature to 8,200 mg COD/L at 13 °C at the same solids retention time. Furthermore, the sludge became bulking at 13 °C with a significant increase in the sludge volume index. The resultant sludge bulking was accompanied by accelerated membrane fouling resulting in a two-fold increase in the frequency of membrane cleaning. The results suggest that performance of MBR activated sludge process deteriorated at low wastewater temperatures even though the effluent water quality was still good enough for its applications in low temperature zones.

Keywords: Low temperature; Membrane bioreactor; Wastewater treatment; Membrane fouling; Nitrifying community structure

Introduction

Due to stringent wastewater discharge regulations and greater need for wastewater reuse, the membrane bioreactor (MBR) activated sludge process has been increasingly used for wastewater treatment. The submerged MBR configuration has been evaluated extensively in terms of its wastewater treatment performance.[1-4] Such a system has the potential to generate high quality effluents with low sludge production[5, 6] and reduced plant footprint[2, 7, 8] because of high biomass concentration operation and excellent solid-liquid separation. With the steady decrease in membrane price and energy consumption,[9, 10] it is expected that MBR systems will be used more and more for wastewater treatment and water reuse.[2, 11]

Many factors affect MBR performance. These include reactor configuration,[1, 12] wastewater composition,[13-15] ambient conditions,[16-18] and important operating parameters such as solids retention time (SRT), hydraulic retention time (HRT),[19, 20] with more details available in recent reviews.[21-23] For instance, dissolved oxygen (DO) level and pH of the mixed liquor may have significant effects on MBR operation. This is because the removal efficiency of soluble microbial products (SMPs), a major cause of fouling in MBR operation[24], decreased with decreasing DO level.[16] A shorter HRT or higher organic loading rate (OLR) can increase membrane fouling rate while membrane fouling rate may decrease when SRT increases.[25-27]

The impact of low temperature operation has been investigated.[7, 28-30] Previous studies have shown that MBR is not sensitive to low temperatures with respect to organic matter removal[31, 32] because of high-density activated sludge operation.[17, 18, 33] Nitrifiers responsible for ammonium removal, however, are very sensitive to temperature changes.[34-36] Nitrification can be significantly inhibited at wastewater temperatures lower than 10 °C and cease completely when the temperature drops below 5 °C.[17, 33, 37] Studies have shown that nitrogen removal efficiency

decreased by more than 60% as the temperature decreased from about 25 °C to 13 °C in submerged MBRs.[33, 38]

Temperature changes affect the bacterial community structure of the activated sludge in MBR as well. For instance, within a wastewater temperature range of 9 °C to 10 °C α-Proteobacteria and certain filamentous bacteria became relatively abundant in the MBR while Proteobacteria, Nitrospirae and Bacteroidetes were the predominant phyla at higher temperatures.[39] Another study found that the dominant bacterial groups in MBR were γ-Proteobacteria, β-Proteobacteria and Nitrospira, and β-Proteobacteria at wastewater temperatures of 30 °C, 20 °C, and 10 °C, respectively.[40] Nitrifying bacteria are essential to nitrogen removal from wastewater. Although they were studied in MBRs at room temperature[25, 41] and at wastewater temperatures ranging of 18 °C to 25 ° C,[42-44] the changes in composition and population abundance of nitrifying bacteria at low wastewater temperatures are not as well understood. It is not clear to which extent the functional redundancy of nitrifiers in the MBR alleviates the adverse effect of low temperature exposure.

Low temperature operation could also accelerate membrane fouling.[3, 33, 40] Although the fouling mechanisms at low temperatures remain to be explored, factors such as release of extracellular polymeric substances (EPS)[39, 45] and increased SMPs[46-48] are believed to be relevant to membrane fouling. Other factors may contribute to membrane fouling at low wastewater temperatures as well. These include increased sludge viscosity, reduced sludge stabilization or sludge deflocculation,[26, 49] reduced particle size of the mixed liquor,[50] and reduced mass transfer efficiency [30]. Whether the fouling of MBR is correlated with sludge bulking at low wastewater temperature operation is, however, largely unknown.

Notwithstanding considerable effort in MBR research and the reports that the MBR process fails at wastewater temperatures lower than 10 °C,[18, 33] the performance of MBR for wastewater treatment in cold climate zones where year-round water temperatures are slightly higher than 10 °C is still poorly studied. As a result, a wastewater temperature of 13 °C was chosen in this study because it is a representative water temperature in many areas in the winter.[3, 46] The objectives of the present study were: 1) to determine the effect of low temperature (~ 13 °C) operation on MBR wastewater treatment performance and activated sludge properties (e.g., biomass concentration, sludge settleability, and nitrifying community structure), and 2) to determine the effect of low temperature operation on membrane fouling.

Materials and methods

MBR operation and monitoring

The MBR was operated as a Modified Ludzack-Ettinger (MLE) system as described previously.[51] Briefly, the MBR with a total working volume of 7.2 L was divided by a plastic baffle into an anoxic chamber (1.8 L) and an aerobic chamber (5.4 L). The system was operated at a HRT of 12 h and a target SRT of 145 d in order to maintain a relatively constant biomass concentration of about 9,000 mg COD/L at room temperature (22 ± 1) °C and the wastewater temperature of (21.5 ± 0.3) °C. The mixed liquor in the aerobic chamber was recirculated to the anoxic chamber at the flow rate that equaled to the influent flow rate. A polyvinylidene fluoride (PVDF) hollow fiber membrane module (ZeeWeed®-1, GE Water & Process Technologies, Trevose, PA) with an effective filtration area of 470 cm^2 and a nominal pore size of 100 nm was submerged in the aerobic chamber for solid-liquid separation. To support bacterial growth and reduce membrane fouling, coarse aeration was applied to the aerobic chamber through the

orifices located at the bottom of the membrane module at a constant flow rate of 9.4 L/min. The water level in the MBR was kept relatively constant (with water volume change < 5%) by using a two-level (upper and lower) sensor (Cole-Palmer, Vernon Hills, IL) while a periplastic pump was operated intermittently after setting the target permeate/effluent flow rate to three times the influent flow rate. The transmembrane pressure (TMP) as an indicator of membrane fouling was monitored daily by a digital pressure gauge (Cole-Palmer, Vernon Hills, IL) while the permeate flux was maintained constantly at an average value of 38.6 ± 0.4 L/(m^2·h).

Synthetic wastewater that was mainly composed of nonfat dry milk powder was used as a feed solution with an average COD concentration of approximately 500 mg/L.[51-53] Other major components of the synthetic wastewater included 51.7 mg/L of total nitrogen (TN), 30 mg/L of NH_4^+-N, and 6 mg/L of total phosphorus (TP). The macro- and micronutrients in the feed solution contained the following: 31.40 mg/L $MgSO_4$, 11.50 mg/L NH_4Cl, 27.70 mg/L Na_2HPO_4, 10.60 mg/L $CaCl_2$, 1.28 mg/L $FeCl_2$, 3.04 mg/L $MnSO_4$, 1.13 mg/L $(NH_4)_6Mo_7O_{24}$, 0.80 mg/L $CuSO_4$, 0.96 mg/L $ZnSO_4$, and 0.15 mg/L $NiSO_4$.

The MBR was seeded with activated sludge from the Columbia Wastewater Treatment Plant (Columbia, MO). The whole MBR study lasted more than 150 d which included about 70 days of MBR operation at room temperature with the rest of the operation at an average wastewater temperature of (13.2 ± 0.4) °C. The MBR system was considered pseudo-steady state based on the sludge properties and consistent effluent water quality (details shown in Fig. 1-3 below because of operation at high biomass concentrations) after about one month of operation under normal and low temperature conditions, respectively. At low temperature operation, the MBR was placed in a closed polystyrene tank that was filled with ice water.

Effluent water quality and activated sludge property

The water quality constituents such as COD, NH_4^+-N, NO_2^--N, NO_3^--N were monitored weekly according to the standard methods.[54, 55] To determine the activated sludge properties at different temperatures, biomass concentration, sludge volume index (SVI), and bacterial activity were monitored after taking the mixed liquor from the aerobic chamber. Biomass concentration was determined in COD units (mg COD/L), which is directly linked to volatile suspended solids concentration.[56, 57] Briefly, aliquots (1 mL) of mixed liquor were removed from the aerobic chamber and were diluted using DI water to a suitable concentration. SVI was also determined weekly according to the standard methods.[54] Each time, 100 mL (~ 2% of the working volume of the aerobic chamber) of the activated sludge was removed from the MBR for SVI determination. The bacterial activities were determined at room temperature and 13 °C, respectively, through the specific oxygen uptake rate (SOUR) measurements following the procedure described previously.[51, 58]

Nitrifying bacterial community structure

The effect of low temperature operation on nitrifying bacterial community structure was determined by terminal restriction fragment length polymorphism (T-RFLP), following the protocols described elsewhere[59] by targeting both ammonia-oxidizing bacteria (AOB) (*β-Proteobacteria*) and nitrite-oxidizing bacteria (NOB) (*Nitrobacter* and *Nitrospira*). The samples at room temperature were collected 3 d before the temperature change, and the samples at low temperature were collected on day 142 (or 72 d after the temperature change). For DNA extraction, aliquots (0.5 mL) of the activated sludge was removed from the aerobic chamber and centrifuged at 10,000 ×g for ~ 3 min (room temperature). Total genomic DNA was isolated from

the pellet using an UltraClean® Soil DNA Isolation Kit (MO-BIO, Carlsbad, CA),[60, 61] following the manufacturer's manual.

Polymerase chain reactions (PCRs) were performed to amplify 16s rRNA gene fragments from the total genomic DNA. The PCR reactions with a total volume of 50 μL contained (final concentration or amount) 2.0 U of GoTaq® DNA Polymerase (Promega, Madison, MI), 2.5 mM magnesium chloride ($MgCl_2$), 1× Colorless GoTaq® Flex Buffer, 0.25 mM (each) deoxynucleoside triphosphate (dNTP), 400 nM (each) forward and reverse primer (Table S3), and 2.0 μL of 10 times diluted DNA sample. All primers were synthesized by Integrated DNA Technologies, Inc (Coralville, IA). The primer sequences and PCR programs are listed in Table S3. The PCR products were confirmed by gel electrophoresis and purified by a Wizard® SV Gel and PCR Clean-UP System (Promega, Madison, MI), following the manufacturer's manual.

The purified PCR products were digested using restriction enzyme *Msp*I (Promega, Madison, MI). Briefly, 18 μL of purified PCR product, 2 μL of *Msp*I restriction endnuclease, and 2 μL of Buffer B were mixed and incubated in 37 ºC water bath for 3 h.[62, 63] The digested PCR products were diluted 10 times using RNase-Free water and then subject to DNA fragment analysis using a 96-capillary ABI 3730xl DNA Analyzer (Applied Biosystems, Foster City, CA)[64, 65] at the University of Missouri DNA Core Facility (Columbia, MO). T-RFLP profiles were further analyzed using a Peak Scanner™ Software v1.0 (Life Technologies Corporation, Carlsbad, CA)[66, 67] to obtain the electropherograms of nitrifying bacteria in the MBR.

Membrane fouling and control

Membrane fouling is accompanied by as an increase in total transmembrane resistance under a constant permeate flux, which is described in the following equation:

$$J = \frac{\Delta P}{\mu \cdot R_t} \quad (1)$$

where J is the permeate flux, μ is the viscosity of activated sludge, ΔP is the TMP, and R_t is the total hydraulic filtration resistance.[68] Membrane fouling is caused by many factors and among them, EPS is considered as an important one.[69, 70] Thus, to determine the effects of low temperature operation on membrane fouling, the EPS concentrations were determined as the sum of the total polysaccharides and total proteins.[51] Polysaccharide content was determined by phenol-sulfuric acid method with D^+-glucose as a standard[71] and the total protein concentration was determined by ultraviolet multi-wavelength absorptiometry.[72]

Throughout the study, the permeate flux was kept constant [~ 39 L/(m^2·h)] in MBR operation. When the TMP reached 43 kPa, the membrane module was taken out of the MBR for cleaning. The cake layer of the membrane module was first removed by flushing the membrane surface with tap water and then it was soaked in a 0.2% (w/v) sodium hypochlorite (NaClO) to further remove the fouling deposits. The membrane module was cleaned again with tap water before it was put back in service.

Statistical analysis

To assess the statistical significance of the difference in wastewater treatment performance before and after temperature change, an unpaired student's *t*-test was conducted with *p*-values less than 0.05 indicating statistical significance.[55, 73]

Results

Impact of low temperature operation on effluent water quality and activated sludge properties

The concentration profiles of the effluent water quality constituents such as COD and main inorganic nitrogen species (NH_4^+-N and NO_3^--N) are shown in Fig. 1 and 2, respectively. The effluent COD concentration increased significantly ($p < 0.001$) from (10 ± 5) (n = 13) to (25 ± 4) mg/L (n = 12) as the wastewater temperature decreased from room temperature (22 °C) to 13 °C (Table S1). Although statistical analysis showed that the effluent NO_3^--N concentration decreased significantly ($p < 0.001$) under low temperature operation conditions, the effluent NO_3^--N concentrations at room and low temperatures were relatively constant at (34.5 ± 0.3) and (32.8 ± 0.6) mg N/L, respectively. Compared with NO_3^--N, the effluent NO_2^--N concentrations were very low (< 0.2 mg N/L) and did not show a significant change under low temperature operation conditions ($p = 0.19$). The average effluent NH_4^+-N concentration was low throughout the experimental period (< 0.1 mg N/L) (Fig. 2, Table S1).

Biomass concentration decreased significantly from (9,967 ± 874) mg COD/L (n = 13) at room temperature to (8,182 ± 606) mg COD/L (n = 12) at 13 °C ($p < 0.001$) (Fig. 3). Meanwhile, the sludge SVI increased significantly from (102 ± 13) mL/g VSS (n = 13) at room temperature to (146 ± 8) mL/g VSS (n = 12) at 13 °C ($p < 0.001$). This indicated that low temperature operation could cause poor sludge settling or sludge deflocculation, resulting in a potential sludge disposal problem.[74]

Changes in nitrifying bacterial activity and population

The heterotrophic SOUR values of the activated sludge at room and low water temperatures were (1.50 ± 0.19) and (0.40 ± 0.03) g O_2/(g VSS·d), respectively. The autotrophic SOUR values at room and low temperatures were (1.58 ± 0.07) and (0.24 ± 0.01) g O_2/(g VSS·d), respectively. Both heterotrophic and autotrophic bacterial activities were significantly reduced at 13 °C.

Seven activated sludge samples (Table S2) were used for T-RFLP analysis and only the representative T-RFLP profiles were presented (Fig. 4). Fig. 4 showed that the genera of *Nitrosospira* and *Nitrosomonas* were present as AOB. For the six AOB groups (*Nitrosomonas europaea/eutropha* lineage, *Nitrosomonas oligotropha* lineage, *Nitrosomonas cryotolerans*, *Nitrosomonas marina* lineage, *Nitrosomonas communis* lineage, and *Nitrosospira* lineage),[59] *Nitrosospira* lineage [terminal fragment (TF) = 101 bp] and *Nitrosomonas europaea*/*eutropha* lineage (TF = 161 bp) were present with *Nitrosomonas europaea*/*eutropha* lineage to be the main AOB as indicated from their very small peak heights (Fig. 4). Other AOB lineages had lower abundance in the activated sludge. Both *Nitrobacter* (TF = 136 bp, data not shown) and *Nitrospira* (TF = 130 bp, 261 bp, and 272/273 bp) were identified as NOB. Based on the peak heights, *Nitrospira* had higher abundance than *Nitrobacter* in the activated sludge.

At low temperature operation, the populations of nitrifiers such as those of the *Nitrosospira* lineage with TF of 101 bp and *Nitrospira* with TFs of 130 bp and 261 bp decreased significantly. On the other hand, the populations of dominant nitrifying species, such as the *Nitrosomonas europaea*/*eutropha* lineage with terminal fragment (TF) of 161 bp and *Nitrospira* with TF of 272/273 bp were almost constant, indicating that these species were not very sensitive to temperature drop. Although less abundant, the population of *Nitrobacter* was relatively constantly at room and low wastewater temperatures (data not shown).

Membrane fouling of the MBR at low temperature operation

The TMP gradually increased with operating time due to membrane fouling while an almost constant permeate flux was maintained in the MBR (Fig. 5). Under room temperature conditions the membrane module required cleaning every 30 days. However, at 13 °C the membrane module required a shorter period of time (< 15 days) to reach the threshold TMP (~ 43 kPa) with at least a two-fold increase in the frequency of membrane cleaning, suggesting accelerated membrane fouling [50]. Meanwhile, the EPS concentrations at room and low wastewater temperatures were (18.5 ± 1.3) and (15.3 ± 1.3) mg/g VSS, respectively (Fig. 6).

Discussion

The permeate or effluent water quality data suggest that low temperature operation resulted in a deterioration of MBR wastewater treatment performance. The results are consistent with other studies,[38] showing that the average effluent COD concentration increased significantly at low temperature operation. Bacteria with lower activity at low temperature operation are susceptible to inhibition and environmental changes, resulting in a decreased organic matter removal. Quantitatively, the effluent COD concentration is defined by the intrinsic kinetic parameters associated with bacterial growth:

$$S_S = \frac{K_s(1/\theta_c + b)}{\mu_{\max} - (1/\theta_c + b)} \quad (2)$$

where S_S is the effluent organic matter concentration of the MBR (mg COD/L), K_S is the half-saturation coefficient (mg COD/L), μ_{max} is the maximum specific growth rate of the heterotrophic bacteria in activated sludge (d^{-1}), θ_c represents SRT (d), and b is the specific heterotrophic decay rate constant. As wastewater temperature decreases, bacteria would have

much higher K_S values (or much lower affinities for substrates)[17, 37] because low temperatures decrease the nutrient transport efficiency of cell membrane proteins.[75] Moreover, due to the limit of nutrient supply at low wastewater temperatures, the maximum specific growth rate μ_{max} would also decrease.[75] As a result, low wastewater temperatures resulted in high effluent COD concentrations or low COD removal efficiencies. The deterioration of effluent water quality might be also linked to the poor compressibility and settleability of activated sludge (indicated by higher SVI values)[76] at low temperature operation. However, the effect of sludge compressibility and settleability on MBR performance would be limited because of the excellent solid-liquid separation characteristics of membrane.[77]

Fig. 2 demonstrates that inorganic nitrogen removal was not significantly affected by low temperature operation. Consistent with previous studies in the MBR system,[51] effluent NO_2^--N was not detected and NH_4^+-N concentrations were very low throughout the study, indicating complete nitrification. The almost complete nitrification appears to be in conflict with significantly reduced autotrophic bacterial activities at low temperature operation, which could be explained in several ways. First of all, the MBR was operated at high biomass concentrations throughout the study. At the wastewater temperature of 13 °C, although the biomass concentration decreased to ~ 8000 mg COD/L, it was much higher than that of a conventional activated sludge process.[17, 37] The high biomass concentration could compensate for the loss of nitrifying activities at 13 °C. Second, nitrifying bacterial communities in activated sludge usually contain a significant amount of functional redundancy,[78] which helps maintain stable nitrification when wastewater temperature drops. Although the populations of some AOB and NOB species decreased, the populations of major AOB with TF of 161 bp (*Nitrosomonas europaea*/*eutropha* lineage) or *Nitrospira* species with TF of 272/273 bp (Fig. 4) were not affected at low

temperature operation. Recently similar results have been reported where *Nitrosomonas* prevailed in the MBR within a wastewater temperature range of 10 °C to 23 °C.[79] Regardless of the temperature change, the MBR system demonstrated its effectiveness in organic removal and complete nitrification because of its operation at high biomass concentrations, which provided a unique niche rich with biodiversity and abundance. As the wastewater temperatures dropped, the microbes that were not sensitive to temperature changes could still achieve high efficiencies of organic matter removal and nitrification. Hence, high biomass concentrations with high microbial biodiversity in the MBR operated at long SRTs could offset the adverse effect of low temperature exposure.

Although the MBR was operated at the same SRT, the biomass concentrations decreased significantly at low temperature operation (Fig. 3). The results were consistent with a previous study in a full-scale MBR where biomass concentrations decreased from summer to winter.[30] This phenomenon can be explained in two ways. First of all, biomass synthesis relies upon the energy released from oxidization of organic matter and/or ammonium. The net energy released from a redox reaction ($-\Delta G$, J/mol e^-) can be expressed in the following equation:[80]

$$-\Delta G = T\Delta S - \Delta H \quad (3)$$

where ΔS is the change in entropy of the reaction, and ΔH is the change in enthalpy of the reaction and is considered almost constant, regardless of temperature (T) changes.[80, 81] As the oxidization of organic matter to carbon dioxide and water increases the randomness of the system[82-84] (with a positive ΔS), less energy (Eqn. 3) is available for biomass synthesis at low wastewater temperatures. Furthermore, as wastewater temperature drops, the nutrient transport efficiency decreases significantly[75] and more energy is required for cell metabolism or maintenance. As a result, less energy released from the oxidization of organic matter can be used

for cell synthesis. In other words, the true yield (Y) or the observed yield (Y_{obs}) in the following equation (Eqn. 4)[17, 37] decreases under low temperature operation conditions.

$$X = \frac{\theta_C}{\tau} Y_{obs}(S_{SO} - S_S) \quad (4)$$

where X is the biomass concentration in the MBR (mg biomass COD/L), Y_{obs} is the observed yield of the activated sludge (mg biomass COD/mg COD utilized), S_{SO} is the influent COD concentration, and τ is HRT. Second, as wastewater temperature dropped from 22 °C to 13 °C, an increase in effluent COD (Fig. 1) and therefore a smaller concentration difference (S_{SO} - S_S) could also contribute to reduced activated sludge concentration at low temperature operation.

This study also demonstrated that low temperature operation resulted in accelerated membrane fouling (Fig. 5), which was consistent with previous MBR studies.[16, 29, 46, 85] Membrane fouling can be grouped into 1) biofouling, 2) organic fouling, and 3) inorganic fouling,[22] where biofouling that is related to EPS and SMP production[86, 87] is considered to be one of the most important factors affecting membrane fouling.[86, 88] Higher EPS and SMP concentrations often resulted in more significant fouling. Here, however, the acceleration of membrane fouling was accompanied by decrease in EPS concentration at low temperature operation. Due to the complexity of fouling mechanisms,[45] many other factors associated with low temperature operation could be therefore more important. First, the sludge SVI values were higher at low operating temperature, indicating poor activated sludge compressibility and settleability.[89, 90] The mixed liquor could have loose morphology and release more small particles at low wastewater temperature.[50] It is known that small sludge particles cause membrane fouling more easily than larger ones.[91] As a result, the TMP increased faster under low temperature operation conditions. Consistent with the fact that there was no correlation between sludge SVI

and EPS concentration,[92] in this study SVI increased while EPS concentration decreased at low wastewater temperatures. Second, unlike normal activated sludge, the bulking sludge might generate more sludge flocs with irregular shape and create a more dense cake layer on the membrane surface,[93] resulting in more significant membrane fouling.[94] Third, as wastewater temperature decreases, the viscosity of the mixed liquor in the MBR would increase,[95] resulting in an increase of TMP at a constant permeate flux (Eqn. 1). Fourth, the higher effluent COD values in the MBR at low temperature operation could contribute to fouling as well.[50] Other factors may also contribute to accelerated membrane fouling at low wastewater temperatures, such as the reduced shear stress generated by air bubbling,[96] low particle back transport velocity,[96] and high hydrophobicity of the activated sludge[89] at low temperature operation.

This study revealed that MBR performance deteriorated at the wastewater temperature of 13 °C. However, for municipal wastewater treatment in low temperature zones, MBR is still a good option for high efficiency COD removal and year-round nitrification. Further research is needed to understand the MBR performance and activated sludge characteristics at lower wastewater temperatures.

Conclusions

This study investigated the effect of low temperature operation on MBR wastewater treatment performance and activated sludge properties. The effluent water quality deteriorated as the COD concentration increased from an average of 10 mg/L at room temperature to 25 mg/L at 13 °C. Although the effluent nitrogen concentrations were not affected under low temperature exposure, nitrifying activity and abundance of nitrifiers decreased significantly at 13 °C. The low temperature operation also resulted in accelerated membrane fouling as revealed by a two-fold

increase in the frequency of membrane cleaning. Nevertheless, the effluent water quality of the MBR was still good, demonstrating the practicality of its use in low temperature zones.

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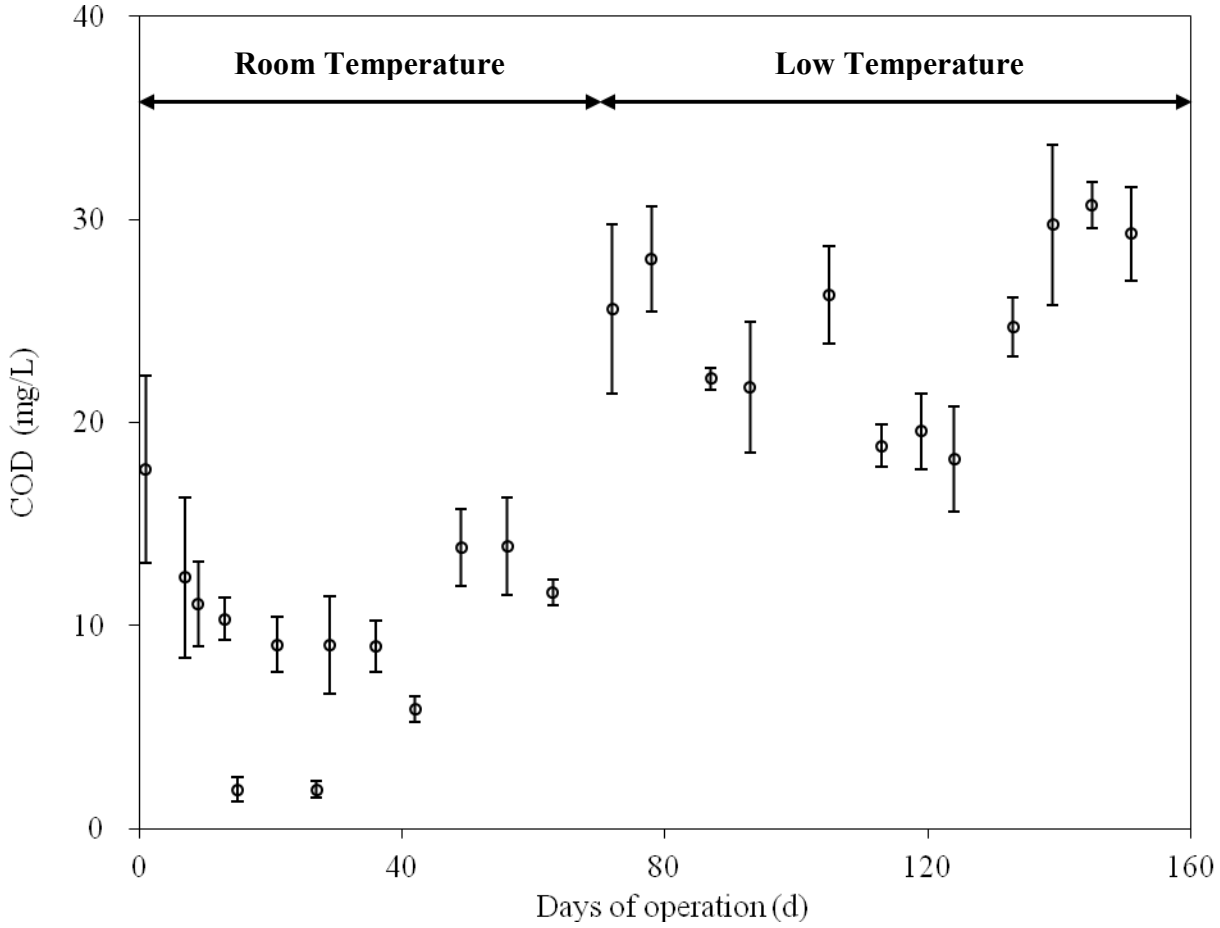

Fig. 1 - Effluent COD concentration at room and low wastewater temperatures. The error bars represent the range of duplicate measurements.

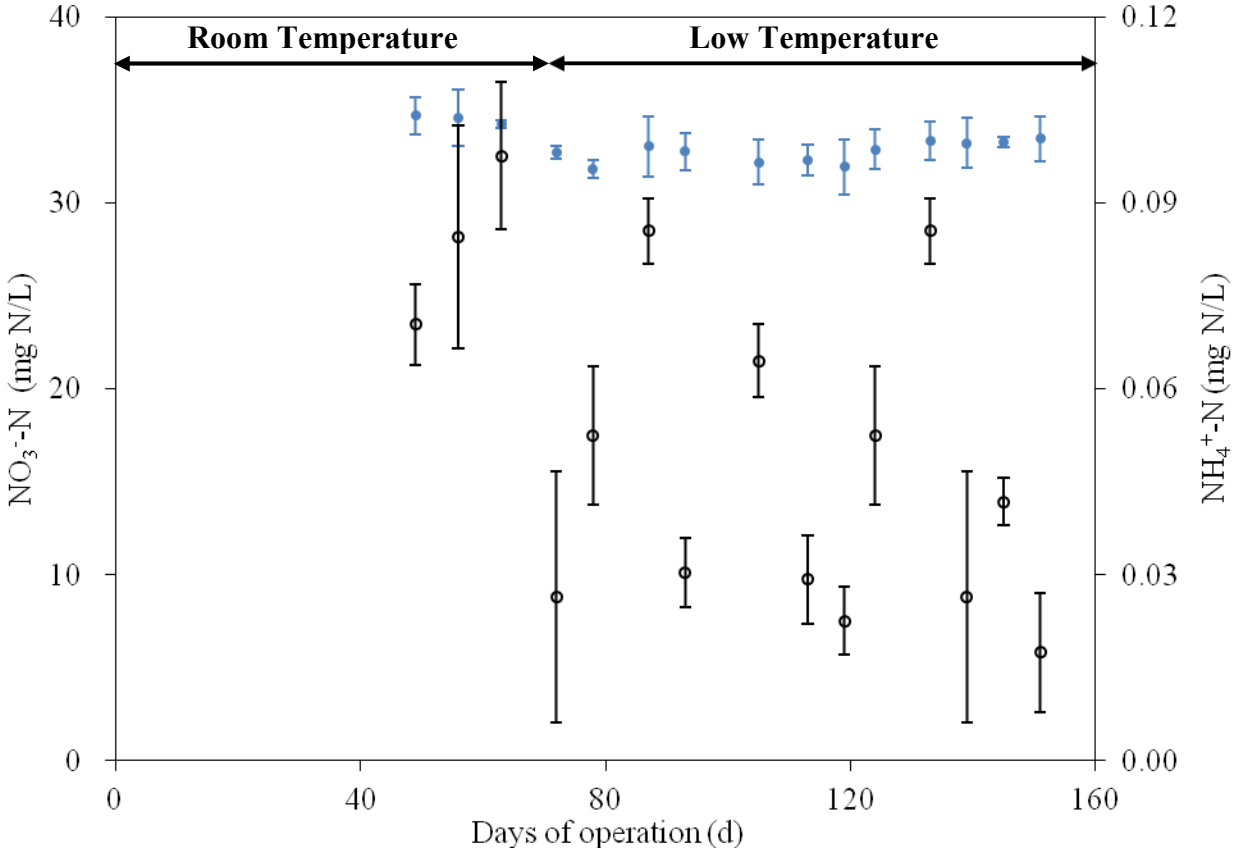

Fig. 2 - Effluent nitrate-nitrogen (●) and ammonium-nitrogen (○) concentrations at room and low wastewater temperatures. The error bars represent the range of duplicate measurements.

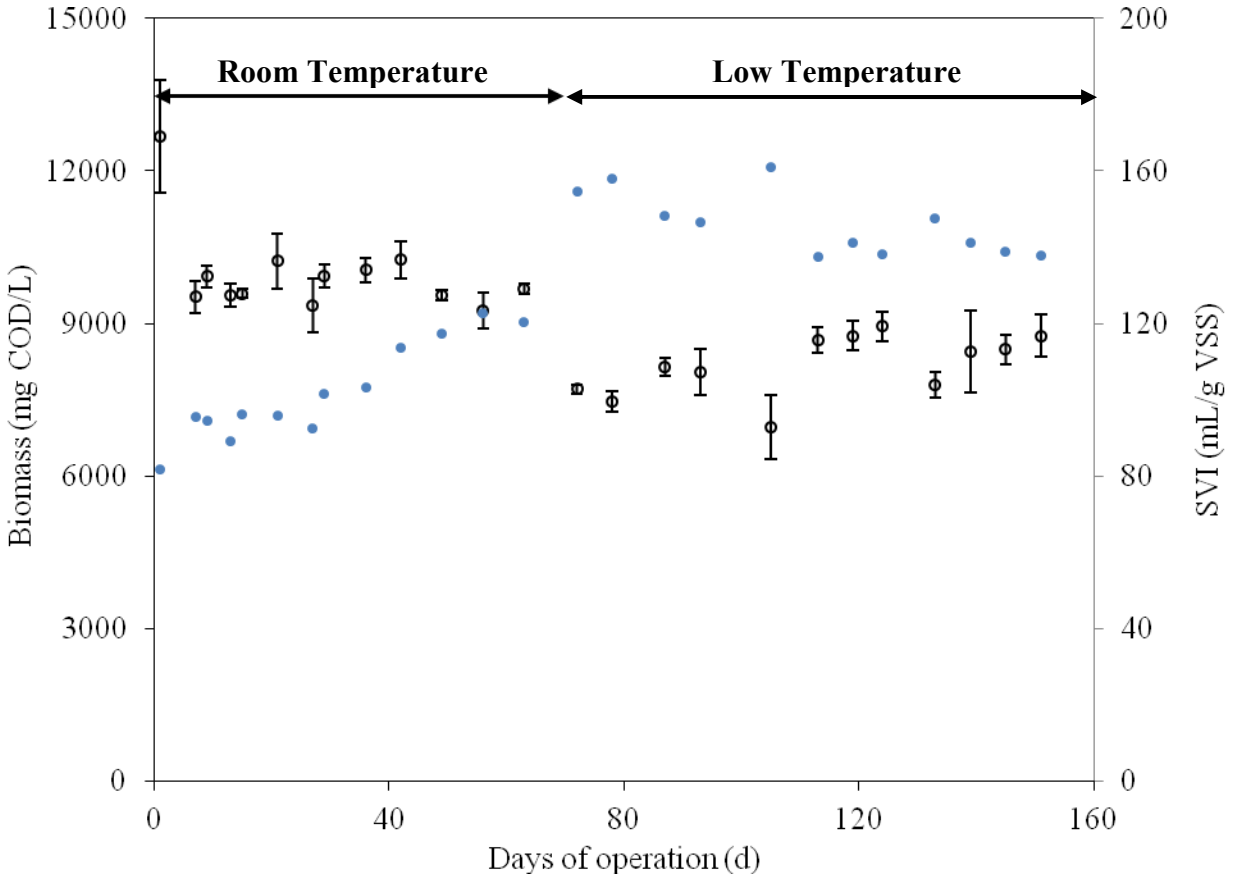

Fig. 3 - Activated sludge biomass concentration (○) and SVI (●) at room and low wastewater temperatures. The error bars represent the range of duplicate measurements.

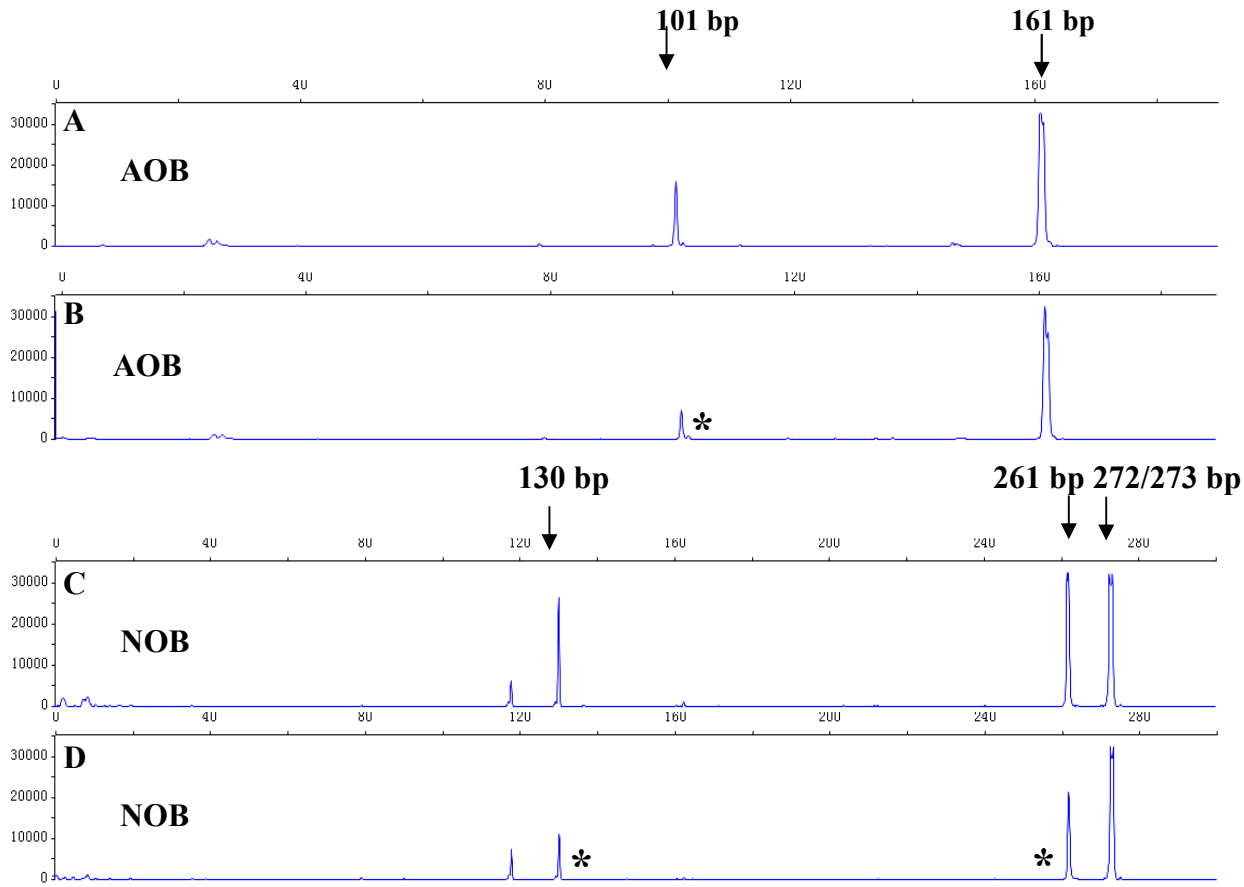

Fig. 4 - Electropherograms of the T-RFLP of nitrifiers at room (A and C) and low water (B and D) temperatures. The room temperature samples (A and C) were collected 3 d before the temperature change, and the low temperature samples (B and D) were collected on day 142 (or 72 d after the temperature change). A and B: T-RFLP results for the β-Proteobacteria AOB group. Arrows correspond to T-RFLPs of AOB: 101 bp for the *Nitrosospira* lineage, and 161 bp for the AOB *Nitrosomonas europaea*/*eutropha* lineage. C and D: T-RFLP results of NOB dominated by *Nitrospira*. Arrows correspond to *Nitrospira* species with TFs at 130 bp, 261 bp, and 272/273 bp. Asterisks (∗) indicate the corresponding nitrifying bacterial population decreased significantly at low temperature operation.

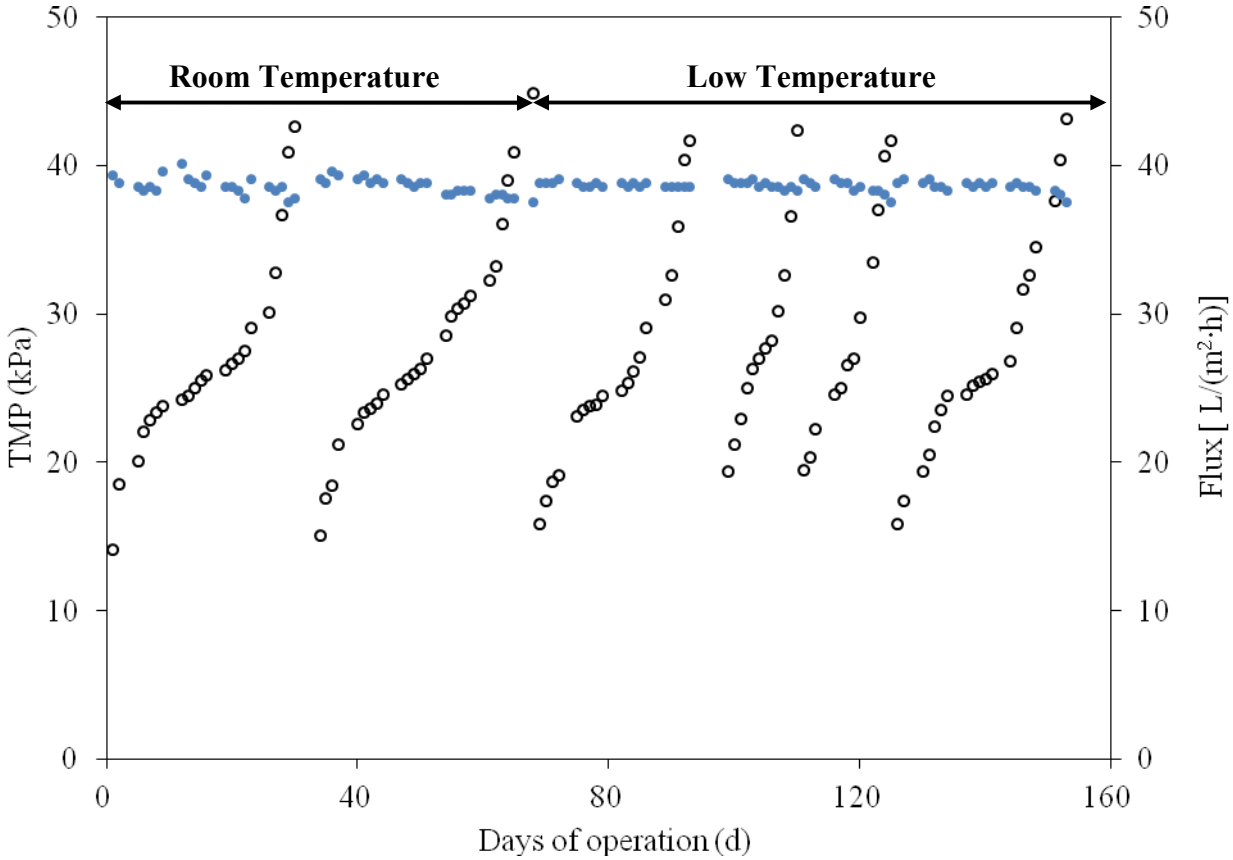

Fig. 5 - TMP (○) and flux (●) of the membrane module at room and low wastewater temperatures.

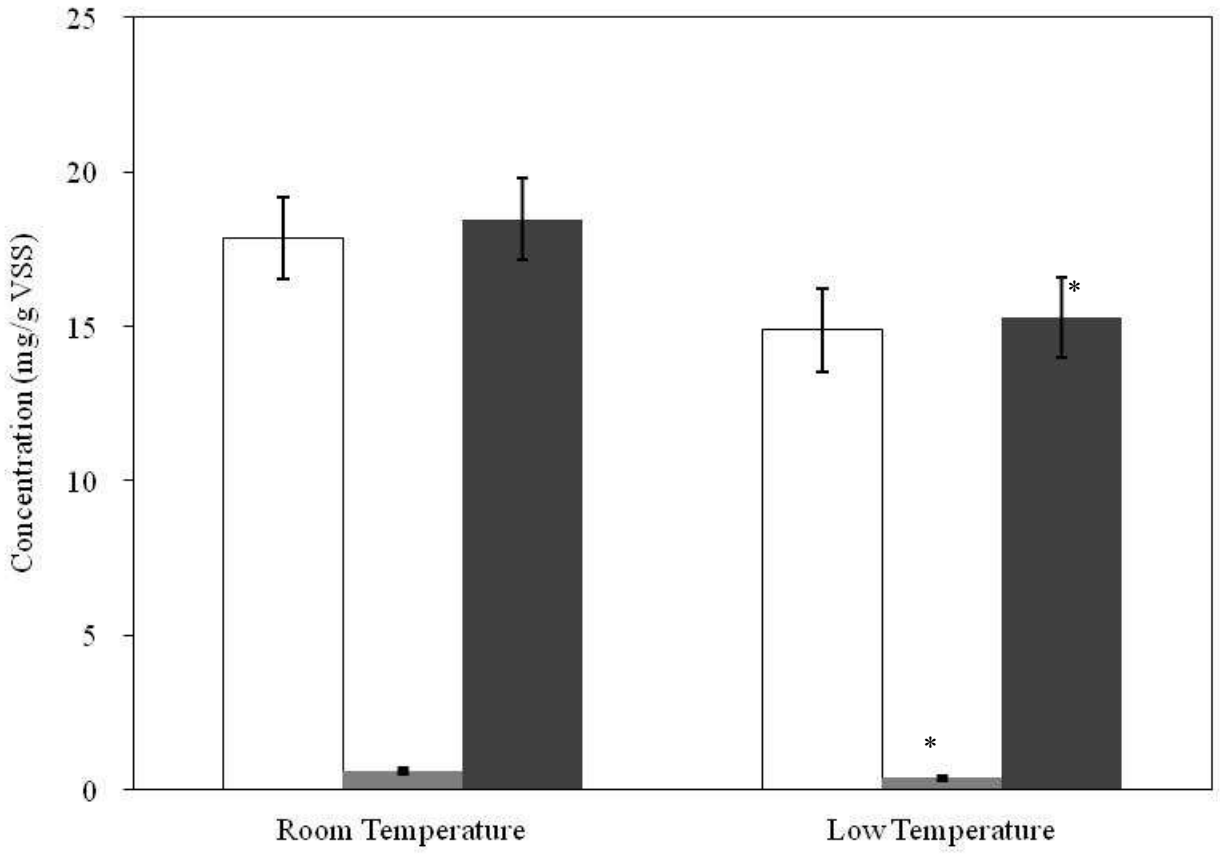

Fig. 6 - Concentrations of EPS (■) including polysaccharide (□) and total protein (■) at room and low wastewater temperatures. Error bars represent the standard deviations (n = 3). Asterisks (*) indicate the concentration decreased significantly under low temperature operation conditions.