



## Research article

## Bacterial community structure and removal performances in IFAS-MBRs: A pilot plant case study



Giorgio Mannina<sup>a</sup>, Marco Capodici<sup>a</sup>, Alida Cosenza<sup>a</sup>, Paolo Cinà<sup>b</sup>, Daniele Di Trapani<sup>a,\*</sup>, Anna Maria Puglia<sup>b</sup>, George A. Ekama<sup>c</sup>

<sup>a</sup> Dipartimento di Ingegneria Civile, Ambientale, Aerospaziale, dei Materiali, Università di Palermo, Viale delle Scienze, Ed. 8, 90100, Palermo, Italy

<sup>b</sup> Dipartimento di Scienze e Tecnologie Biologiche Chimiche e Farmaceutiche, Università di Palermo, Viale delle Scienze, Ed. 16, 90100, Palermo, Italy

<sup>c</sup> Water Research Group, Department of Civil Engineering, University of Cape Town, Rondebosch, 7700, Cape, South Africa

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## ABSTRACT

The paper reports the results of an experimental campaign carried out on a University of Cape Town (UCT) integrated fixed-film activated sludge (IFAS) membrane bioreactor (MBR) pilot plant. The pilot plant was analysed in terms of chemical oxygen demand (COD) and nutrients removal, kinetic/stoichiometric parameters, membrane fouling and sludge dewaterability. Moreover, the cultivable bacterial community structure was also analysed.

The pilot plant showed excellent COD removal efficiency throughout experiments, with average value higher than 98%, despite the slight variations of the influent wastewater. The achieved nitrification efficiency was close to 98% for most of the experiments, suggesting that the biofilm in the aerobic compartment might have sustained the complete nitrification of the influent ammonia, even for concentrations higher than 100 mg L<sup>-1</sup>. The irreversible resistance due to superficial cake deposition was the mechanism that mostly affected the membrane fouling. Moreover, it was noticed an increase of the resistance due pore blocking likely due to the increase of the EPS<sub>Bound</sub> fraction that could derive by biofilm detachment.

The bacterial strains isolated from aerobic tank are wastewater bacteria known for exhibiting efficient heterotrophic nitrification–aerobic denitrification and producing biofilm.

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## 1. Introduction

Nutrients (particularly nitrogen, N, and phosphorus, P, compounds) may have adverse environmental impacts when discharged at high concentration (e.g., eutrophication, toxicity towards the aquatic organisms, etc.) (Wang et al., 2006). Therefore, nutrients abatement is an imperative requirement when discharging in sensitive areas (Li et al., 2013). In the last years, several biological and physic-chemical methods have been applied for nutrients removal from wastewater. Among these methods, biological treatments are the most cost-effective ones (Chu and Wang, 2011). Several investigations has been carried out in order to get useful insight on biological nutrient removal (BNR) (*inter alia* Wanner et al., 1992; Cosenza et al., 2013; Lu et al., 2015). BNR is operated thanks to the activity of autotrophic bacteria,

heterotrophic bacteria and polyphosphate-accumulating organisms (PAOs), under alternating anaerobic/anoxic/aerobic conditions (Naessens et al., 2012; Li et al., 2013). Traditionally, nitrogen removal is accomplished by the joint activity of ammonia oxidizing bacteria (AOB) and nitrite oxidizing bacteria (NOB) for nitrification, while heterotrophic organisms are responsible of denitrification. Biological phosphorus removal exploits PAOs ability to accumulate P as intracellular polyphosphate under the alternation of anaerobic/aerobic conditions. Although activated sludge are effective for removal of organic and nutrient compounds, the overall efficiency is strictly related to the performance of the solid-liquid separation phase into the final settler, which may suffer of separation problems (Wanner, 2002). In this context, membrane bioreactors (MBRs) may represent a useful solution, since they enable to disconnect the efficiency of the biological processes from the biomass settling properties. In particular, MBRs generally feature high quality effluent, small footprint and low sludge production rates compared to conventional activated sludge (CAS) systems

\* Corresponding author.

E-mail address: [daniele.ditrapani@unipa.it](mailto:daniele.ditrapani@unipa.it) (D. Di Trapani).

(Stephenson et al., 2000; Mannina and Di Bella, 2012). Therefore, in the last years the integration of BNR process with MBRs has been proposed for wastewater treatment, in order to increase the effluent quality, including such BNR processes as University of Cape Town (UCT) process, anoxic/oxic (A/O) process and anaerobic/anoxic/oxic (A2O) process (Hu et al., 2014). One of the major drawbacks in MBRs is still represented by fouling phenomena that may severely affect the filtration properties of the membrane modules (Judd and Judd, 2010). Particularly, the mixed liquor suspended solid (MLSS) concentration has been recognized to play a significant effect on membrane fouling (Poyatos et al., 2008; Di Trapani et al., 2014). An alternative to managing this problem is to couple a MBR system with a moving bed biofilm reactor (MBBR) in an IFAS mode (Integrated Fixed Film Activated Sludge). In this way, it enhances the simultaneous growth of suspended biomass and biofilm inside the system (Mannina et al., 2011), realizing a so-called moving bed membrane bioreactor (MBMBR) or MBBR-based integrated IFAS membrane bioreactor (IFAS-MBR) (Leyva-Díaz et al., 2013; Yang et al., 2014; Mannina et al., 2017). In particular, MBBR processes rely on the use of small plastic carrier elements that are kept in constant motion throughout the entire volume of the reactor, for biofilm growth (Ødegaard, 2006; Di Trapani et al., 2008, 2010). These systems are especially useful when slowly growing organisms as nitrifiers have to be maintained inside a wastewater treatment plant (WWTP) (Kermani et al., 2008). When combined with a MBR system realizing a MBMBR process, there is the potential to utilize best characteristics of both biofilm processes and membrane separation (Ivanovic and Leiknes, 2008). Using this technology, the biofilm system may reduce the concentration of suspended solids thus reducing the extent of membrane fouling. Nevertheless, IFAS-MBRs are relatively new, especially when referring to system performance, biomass biokinetic activity and membrane fouling tendency. Moreover, very few studies have been reported for BNR systems adopting hybrid IFAS-MBR processes (Yang et al., 2010). Literature reports that the successful of MBBR in carbon and nutrient removing is related to the cultivated bacteria community and functionality of the organisms that grow on the surface of the carriers and to their competition with suspended organisms (Biswas et al., 2014). Therefore, to better understand the performance of hybrid IFAS-MBR processes, it is necessary to study the microbial communities structure and enzyme activities both for biofilm and suspended organisms.

The aim of the present study is to gain insight about the behaviour of a University of Cape Town (UCT) pilot plant, combining both MBR and MBBR technology (UCT-IFAS-MBR), for the treatment of domestic wastewater. In particular, a UCT-IFAS-MBR pilot plant was monitored for two months without sludge withdrawals with the aim to investigate the system performance in terms of organic carbon and nutrients removal, biomass biokinetic behaviour, membrane fouling tendency and sludge features. Furthermore, the microbial communities' structure was also evaluated during experiments. The derived results are relevant also in view of the development of mathematical modelling tools for design and manage such systems (Freni et al., 2009; Mannina and Viviani, 2009).

## 2. Material and methods

### 2.1. UCT-IFAS-MBR system description

Three in-series reactors, anaerobic, anoxic and aerobic (volume equal to 62 L, 102 L and 211 L, respectively) were realized in accordance with UCT lay-out (Ekama et al., 1983). An aerated MBR reactor (36 L) contained the ultrafiltration hollow fibre membrane (PURON<sup>®</sup>, courtesy of Koch Membrane Systems, Inc.). The mixed

liquor recycled from the MBR to the anoxic reactor ( $Q_{RAS}$ ) was previously conveyed to a 40 L oxygen depletion reactor (ODR). The permeate extraction was replaced each 9 min by a minute of backwashing pumping a small volume of permeate through the membrane module from the Clean In Place (CIP) compartment. From the volumes of the reactors and recycle flows the mass fractions of the reactors were calculated from Ramphao et al. (2005) to be: anaerobic 0.071, anoxic 0.232, aerobic 0.481, MBR + ODR 0.216. The anoxic and aerobic compartments were filled with suspended plastic carriers (courtesy of Amitec Co. Ltd, carriers density =  $0.95 \text{ g cm}^{-3}$ ; carriers specific surface =  $500 \text{ m}^2 \text{ m}^{-3}$ ), with a 15 and 40% filling fraction, corresponding to a net surface area of 75 and  $200 \text{ m}^2 \text{ m}^{-3}$  in the anoxic and aerobic reactor, respectively. For the schematic layout of the UCT-IFAS-MBR pilot plant, the reader is referred to the literature (Mannina et al., 2017).

The permeate net flow rate was set equal to the inlet flow rate  $20 \text{ L h}^{-1}$  ( $Q_{IN}$ ). In compliance with the UCT scheme, the recycle flows were set as follows:  $Q_{R1} = Q_{IN}$  from the anoxic to the anaerobic reactor;  $Q_{R2} = 5 \cdot Q_{IN}$  from the aerobic to the MBR reactor;  $Q_{RAS} = 4 \cdot Q_{IN}$  from the MBR to the anoxic reactor.

The UCT-IFAS-MBR pilot plant was operated for 60 days and was fed with a mixture of real wastewater (deriving from the University buildings and characterized by higher ammonia content compared to typical domestic wastewater) and synthetic wastewater. The latter represented almost 50% of the total wastewater in terms of COD, with the 30% constituted by readily biodegradable COD (RBCOD) (dosed as sodium acetate), whilst the remaining 70% was more slowly biodegradable (dosed as glycerol). The synthetic wastewater was spiked to meet the design organic loading rate to the pilot plant.

The inlet wastewater had the following average features: COD =  $607 \text{ mg L}^{-1}$ ; total nitrogen (TN) =  $65 \text{ mg L}^{-1}$ ; total phosphorus (TP) =  $11 \text{ mg L}^{-1}$  with a COD/TN/TP ratio of 100/10.7/1.8. The permeate flux was maintained equal to  $21 \text{ L m}^{-2} \text{ h}^{-1}$ , whereas the hydraulic retention time (HRT) was equal to 20 h. The pilot plant was operated without sludge wasting during experiments; therefore, it was characterized by an increasing value (indefinite) of the mixed liquor sludge retention time (SRT). The average value of the liquid temperature (T) and pH were  $25 \text{ }^\circ\text{C}$  and 7.8, respectively. The dissolved oxygen (DO) concentrations (average values) ranged from  $0.03 \text{ mg L}^{-1}$  in the anaerobic compartment to 5.33 in the MBR compartment, respectively.

### 2.2. Analytical methods

During experiments, samples from inlet wastewater, permeate and from each reactor were collected in order to analyse total suspended solid (TSS) as well as volatile suspended solids (VSS) concentrations. Organic carbon content was assessed by total chemical oxygen demand ( $\text{COD}_{TOT}$ ), and supernatant COD ( $\text{COD}_{SUP}$ ). Nitrogen forms (ammonium, nitrite and nitrate) and total nitrogen (TN) were assessed as well as orthophosphate and total phosphorus (TP). For further information regarding the analytical methods applied, the reader is addressed to Standard Methods (APHA, 2005). A multi-parameter probe provided the dissolved oxygen (DO) concentration as well as pH value in each reactor. Referring to the COD removal, both the biological removal and the total removal (after membrane filtration) were assessed according to Mannina et al. (2016a). Periodic tests on carrier samples were carried out, in order to evaluate the biofilm growth, in accordance to previously reported methods (Di Trapani et al., 2014).

Biokinetic parameters were measured by means of respirometric batch test carried out in accordance with Di Trapani et al. (2015). Moreover, ammonium and nitrate utilization rate (AUR and NUR respectively) were assessed by applying a modified

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