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Production and Use of Short Chain Fatty Acids to Enhance the Via-Nitrite Biological Nutrients Removal from Anaerobic Supernatant

Nicola Frison[a], Letizia Zanetti[b,c], Evina Katsou[b,c], Simos Malamis[b,c], Franco Cecchi[b,c], Francesco Fatone[b,c,*]

[a]Department of Environmental Sciences, University of Venice, Calle Larga Santa Marta, Venice, Italy
[b]Department of Biotechnology, University of Verona, Via Le Grazie 15, 37134, Verona, Italy, francesco.fatone@univr.it
[c]Interuniversity Consortium "Chemistry for the Environment", Via delle Industrie, 30175, Marghera, Venice, Italy
francesco.fatone@univr.it

The use of the different external carbon source was investigated to enhance the short-cut nitrification-denitrification (SCND) and the denitrifying phosphorus removal via nitrite (DPRN) using a sequencing batch reactor (SBR) to treat anaerobic supernatant. The SBR was fed with liquid effluent from the anaerobic co-digestion of waste activated sludge (WAS) and organic fraction of municipal solid waste (OFMSW). After the start-up period of 20 days, a stable and reliable via-nitrite pathway was achieved. Then, three available carbon sources (fresh drainage liquid from OFMSW, fermentation liquid from OFMSW, cattle manure and maize silage (CM&MS) fermentation liquid) were investigated. Significant in-situ specific denitritation rates (sNUR) in the range of 0.51-1.16 kgNO_2-N / kgVSSd were obtained for OFMSW and CM&MS related carbon sources. The specific consumptions obtained for the liquid effluents from OFMSW and CM&MS raged from 2.7-3.1 kgCOD / kgNO_2-N_{rem}. The presence of short chain fatty acids (SCFA), namely butyric and propionic acids in the fermentation liquid enhanced the DPRN. The cost of the external carbon source can be lower than 0.1 € / kgCOD when OFMSW is fermented. The total specific cost for nitrogen removal may decrease by 22% when the OFMSW fermentation liquid is used instead of methanol.

1. Introduction

Anaerobic digestion is a sustainable biological process used to recover energy from waste. Taking advantage of the relevant EU legislation that promotes the production of electricity from renewable from energy (Directive 2001/77/EC), anaerobic digestion and co-digestion plants for biogas production have become widespread (De Baere, 2006). The solid stream of the digestate can be further treated to produce high quality compost for land application in agriculture. Nutrients recycling originating from domestic waste could replace 35-45% of the produced fertilizers (Lind et al., 2001). Anaerobic digestion of organic substrates like the organic fraction of municipal solid waste (OFMSW), agricultural by-products, activated sludge results in the solubilisation of nitrogen and phosphorus; consequently the anaerobic supernatant is characterized by significant concentrations of ammonium and phosphates and should therefore receive adequate treatment prior its final disposal or reuse using sustainable techniques (Cervantes, 2009; Zanetti et al., 2012). Biological nitrogen removal via-nitrite has received increasing attention due to its advantages compared to the conventional via nitrate nitrogen removal (Fatone et al., 2011). The oxidation of ammonium to nitrite and its reduction to nitrogen gas requires less external carbon source than the conventional nitrification/denitrification. The via nitrite nitrogen removal pathway theoretically decreases the oxygen and the external carbon source requirements up to 25% and 40% respectively compared to the via nitrite pathway. In addition, it reduces the production of sludge and CO_2 emissions by 30% and 20%

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respectively (Gustavsson, 2010). The price of commercial synthetic carbon sources has greatly increased over the last years (e.g. methanol cost in Europe is currently 370 €/t (www.methanex.com), thus promoting the use of non-conventional carbon sources. Significant cost savings can arise when liquid effluents produced from biowaste are utilized as external carbon source (Mayer et al., 2009; Ji and Chen, 2010). Short chain carbon sources produced during the fermentation process can enhance denitritation and denitrifying phosphorus removal via nitrite (DPRN) from the anaerobic supernatant (Frison et al., 2013). Currently, the use of external short chain carbon sources from fermentation liquids is the most common and effective practice. Mayer et al. (2009) used mashed liquid biowaste as an external carbon source for Denitritation in a sequencing batch reactor (SBR) treating anaerobic supernatant. This system achieved significant COD removal (83 %) and high nitrogen removal (95 %). Torà et al. (2011) examined the effectiveness of glycerol, ethanol, landfill leachate, acid-fermented primary and secondary sludge centrate in the denitritation process using two lab scale SBRs. Ji and Chen (2010) used sludge fermentation liquid as carbon source in an SBR treating municipal wastewater in order to enhance SCND and DPRN. The aim of this work was to determine the most effective carbon source that will promote the SCND and DPRN using non conventional liquid effluents produced from bioprocesses.

2. Materials and Methods

2.1 The integrated scheme of Treviso and scSBR (short-cut Sequencing Batch Reactor)

Figure 1 diagrammatically represents the integrated treatment of OFMSW and municipal wastewater in Treviso (Veneto Region).

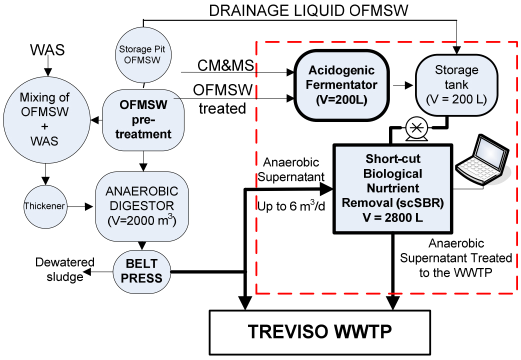

Figure 1: Schematic representation of OFMSW & WAS anaerobic co-digestion and supernatant treatment unit in Treviso WWTP

The anaerobic digester (working volume of 2000 m^3) was fed with a mixture of 120 m^3d^{-1}±30% of thickened waste activated sludge and 5.1±95% td^{-1} of source separated OFMSW, to achieve an organic loading rate of 1.7 kgTVS $m^{-3}d^{-1}$. The hydraulic retention time (HRT) of the anaerobic reactor was 20 days, operating at mesophilic temperature (37°C). The pilot short-cut sequencing batch reactor (scSBR) had a working volume of 2.8 m^3 and operated for 160 d treating up to 6 m^3d^{-1} of anaerobic supernatant of WAS and OFMSW, taken directly from the full scale belt press of the WWTP and stored in a tank of 10 m^3 (Frison et al., 2012). From this tank, the supernatant was pumped from a monho pump into the reactor. The scSBR was equipped with two peristaltic pumps which allowed, when necessary, the dosing of chemicals such as the NaOH solution (30%) to correct the pH and external carbon sources. Aeration was provided by three blowers that were automatically switched on/off through the processing of DO measurements (DO Hach-Lange probe) by a PLC (Telemecanique, Schneider Electric, Germany) with a control algorithm. The software was also able to process and record other analog signals from submerged probes of pH, ORP, NH_4-N, NO_x-N, MLSS and conductivity (Hach-Lange) and temperature. The scSBR worked with an average temperature of 17±2 °C, while the system's heating was set in order to avoid operating temperatures lower than 15°C.

2.2 Origin of the external carbon source to enhance the short-cut biological nutrients removal

Three external carbon sources (fresh drainage liquid from OFMSW piles, OFMSW fermentation liquid, and CM&MS fermentation liquid) were tested through in-situ and ex-situ experiments of denitrification and phosphorus removal via-nitrite. The fresh drainage liquid (DL) was the leachate residual produced during the storage of the raw OFMSW in the full-scale storage pit of the Treviso WWTP. The observed production and the accumulation rate of the DL OFMSW was estimated to be around 100 L per tonne of raw OFMSW; its production depended on the room temperature and the quality of the raw OFMSW. The DL OFMSW was produced from uncontrolled fermentation activities, due to the prolonged retention time in the storage tank. The FL OFMSW and FL CM&MS were obtained in a 200 L pilot reactor, according to the procedure described by Frison et al. (2013). The liquid biowaste carbon sources were collected in a 200 L storage tank and were then dosed to the scSBR.

2.3 In-situ and ex-situ activity test

The three external carbon sources were applied in the last experimental period (days 120-160) of the experimental work carried out by Frison et al. (2013). The phases of the scSBR cycle were the following: 15-17 min feed, 80 min anoxic phase, 160 min aerobic phase, 10 min of discharge 30 min of settling. The overall hydraulic retention time (HRT) was 0.52 d and the sludge retention time was higher than 20 d. The average pH in the effluent was 7.88, in order to avoid inhibiting effects from the production of free nitrous acid (FNA<0.005 mgN/L), while the concentration of the free ammonia (FA) in the effluent varied from 1.26 to 10.03 $mgNH_3$-N/L. The carbon sources were dosed instantly at the beginning of the anoxic phases (OLR = 2.13 $kgCOD/m^{-3}d$), which were studied monitoring in real-time the soluble nitrogen forms (NO_2-N, NO_3-N) and phosphate (PO_4-P) in order to determine the specific nitrite uptake rate (sNUR) and the specific phosphorus uptake rate (sPUR). The in-situ activities tests of the biomass, in terms of nitrite and phosphate removal rate, were carried out in mixed liquor samples collected from the scSBR every ten minutes during all the cycles. During the experimental period, three ex-situ experiments (each for carbon source) were also carried out together with the in-situ monitoring in order to investigate the nitrite and phosphorus removal under anoxic conditions, through the dosage of different external carbon sources. Each batch reactor was composed of a stirred glass vessel with two litres of activated sludge, which was taken directly from the scSBR at the end of aerobic phase, typically with PO_4-P/NO_2-N ratio varying from 0.18-0.24 $kgPO_4$-P/$kgNO_2$-N, in order to avoid any addition of nitrite or phosphate solution. The anoxic conditions were ensuring by stripping the residual oxygen by supplying for 10 minutes nitrogen gas in the mixed liquor. The desired mixing in the glass flask was achieved with a magnetic stirrer while the temperature was maintained at 20±1 °C. The external carbon source was dosed during the anoxic phase in order to have an initial COD/PO_4-P ratio in the range of 11-13 kgCOD/$kgPO_4$-P. The samples, taken every 10 minutes, were analysed for the nitrite, nitrate and phosphate concentration, in order to develop the profiles and the maximal removal rates of the nutrients. In order to compare all nitrogen and phosphorus removal rates from in-situ and ex-situ tests, the slope of the profiles were corrected to the reference temperature of 20^oC.

2.4 Analytical methods

The anaerobic supernatant, was sampled from the accumulation tank and analysed twice a week for all nitrogen and phosphorus forms (NH_4-N, NO_2-N, NO_3-N, TKN, TP, PO_4-P), total COD and soluble COD (sCOD) according to standard methods (APHA, AWWA, WEF, 1998) and ion chromatography (Dionex IC-90 with AG14 and AS14 columns). Besides chemical and physical analysis, respirometric investigation was carried out using the MARTINA respirometer (SPES), to determine the soluble and biodegradable carbon species of the external carbon sources. In addition, the quantification and the composition of the Volatile Fatty Acids (VFA) was carried out by gas-chromatography using a Nukol 15 m column, working at 85-125 °C (rate 30 °C/min) and nitrogen gas as carrier (flow 5 ml/min).

3. Results and discussion

3.1 Characteristics of supernatant and of external carbon sources

Table 1 summarizes the physicochemical characteristics of the anaerobic supernatant that was fed to the SBR. The TSS concentration was low indicating that the solid/liquid separation process of the anaerobic effluent was effective. The ammonium nitrogen concentration was high due to the solubilisation of organic nitrogen to ammonia, while the organic matter was very low owing to effective anaerobic decomposition.

A complete physicochemical and respirometric analysis of the carbon sources was conducted to evaluate the chemical forms and the degree of biodegradability of organic matter (Table 2). The selected carbon sources were generated from biowaste and can thus be obtained in full scale anaerobic digestion plants. The available carbon sources are characterized by significant concentration of short chain fatty acids (SCFA) including acetic, propionic and butyric acid (Figure 2).

Table 1: Physicochemical characteristics of the anaerobic supernatant fed to the SBR

Parameter	Average	Variation (min – max)
Total COD (mg/L)	30.9 ± 12.39	21.5 - 56.6
Soluble COD (mg/L)	25.7 ± 21.29	18.6 - 31.9
TN (mg/L)	570.3 ± 107.06	430 - 670
NH_4-N (mg/L)	558.9 ± 39.53	411 - 650
TP (mgP/L)	31.3 ± 3.00	25.6 - 33.6
PO_4-P (mgP/L)	30.1 ± 5.16	12 - 32
Conductivity (ms/cm)	4.44 ± 0.11	3.88 - 5.21
Na (mg/L)	98.2 ± 5.6	
K (mg/L)	82.0 ± 9.8	
Mg (mg/L)	45.8 ± 3.1	
Ca (mg/L)	79.2 ± 9.4	

SCFA can decrease nitrate formation from 24–70% without affecting the activity of AOB in the activated sludge processes (Gomez et al., 2000; Oguz et al., 2006). The type of carbon source can also significantly affect the denitrifying phosphorus removal (Carvalho et al., 2007). The fermentation liquids from OFMSW and MS&CM had a sCOD/COD ratio of ~50%, while the DL OFMSW had a sCOD/COD ratio up to 73%, due to its lower content in organic particulate matter (6 gTS/kg) compared to that of the FL (25 gTS/kg for FL OFMSW and 48 gTS/kg for FL CM&MS). As seen in Table 2, the controlled fermentation did not result in significant changes in the sCOD and rbCOD concentration in the OFMSW carbon sources. The results of the respirometric analysis showed that the ratio of rbCOD/sCOD was higher than 90% for the OFMSW-related carbon source and ~65 % for CM&MS FL. On the other hand, the fermentation process resulted in an increase of SCFAs up to 60 % of the sCOD, while around 16 % of the sCOD was present in the raw drainage of OFMSW piles.

Table 2: COD characterization of each carbon source

Parameter	DL OFMSW Average (Variation)	FL OFMSW Average (Variation)	FL CM&MS Average (Variation)
Total COD (g/L)	58 (52-64)	62 (56-68)	85 (80-90)
sCOD (g/L)	42 (33-51)	31 (28-34)	43 (37-49)
rbCOD (g/L)	38 (29-47)	29 (25-33)	28 (23-33)
TVFA (g/L)	6.9 (6-8)	18.2 (16-20)	26.5 (24-30)
C/N (gCOD/gN)	26 (24-29)	57 (51-62)	17 (16-18)

The acid fermentation resulted in the production of 199 and 273 gCOD/kgTVS SCFAs for OFMSW and CM&MS, respectively. However, the SCFAs in the MS was already high (20-25 COD/$gMS_{wet\ weight}$). As seen in Figure 2 the main SCFAs of the applied carbon sources were acetic, propionic and butyric acid, while the content of acetic acid was always higher than 4.4 gCOD/L. The latter is in agreement with the results of Traverso et al. (2000) who concluded that organic matter in the liquid phase inside a fermenter fed with mixtures of vegetables and fruits mainly consisted of acetate. Other fermentation products are lactic acid and several alcohols, as a result of alcoholic and lactic transformation of pyruvate (Traverso et al., 2000). Biofibres in the OFMSW and in CM&MS are not completely degraded during the acidogenic fermentation. As a result, humic acids with a high molar mass may form and accumulate in the FL up to 26% of the total COD (Tong and Chen, 2009). However, previous research studies have shown that the presence of HA may enhance the nitrogen and phosphorous removal via the nitrite pathway (Tong and Chen, 2009; Ji and Chen, 2010). Due to the ammonification that takes place during the acid fermentation, nutrients solubilisation resulted in an increase of ammonia concentration in the fermented external carbon sources. Thus, the fermentation liquids produced from OFMSW and CM&MS contained 100-382 gsCOD/gNH_4-N and 45gsCOD/gNH_4-N, respectively. Phosphorus ranged from 258-138 gsCOD/gPO_4-P for the fermentation liquids, while 467 gsCOD/gPO4-P was contained in the DL OFMSW.

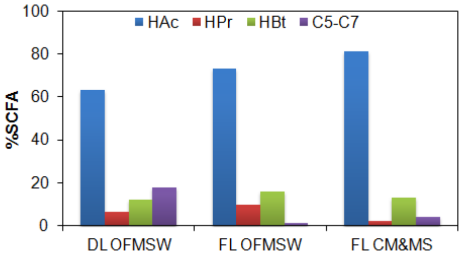

Figure 2: Composition of SCFA in the liquid phase of the carbon sources (HAc=Acetic acid, HPr=Propionic acid, HBt=Butyric acid)

3.2 Effect of external carbon source on nitrogen and phosphorous removal

In this section the effect of the type of external carbon source (i.e. SCFA) on SCND and DPRN was examined. Figure 3a & 3b show the effect of the applied carbon sources on sNUR and sPUR for in-situ and batch experiments respectively. In the in-situ experiments the fermentation liquid of CM&MS resulted in the highest sNUR (i.e.1.16±0.12 kgNO_2–N/ kgVSSd). However, the addition of CM&MS is accompanied by nitrogen overloading, since it significantly increases the NH_4-N content. The ammonium content of FL CM&MS was 0.93 ± 0.14 mgNH_4-N/L, value that is higher than that of DL OFMSW (0.11±0.03 mgNH_4-N/L) and FL OFMSW (0.31±0.01 NH_4-Nmg/L). In the in-situ experiments (Figure 3a), the FL CM&MS resulted in the highest sPUR and sNUR, with the DL OFMSW exhibited the worst performance. The increased sPUR in the FL may be related to the promotion of phosphorus accumulating organisms (PAOs) instead of glycogen accumulating organisms (GAOs) (Ji and Chen, 2010). The results showed that the type of external carbon source critically impacted on the performance of denitrifying bacteria. In batch experiments, the highest sNUR and sPUR were obtained for the FL OFMSW (Figure 3b), while DL OFMSW resulted in the lowest sPUR. FL OFMSW contains much more readily available SCFAs than the DL OFMSW, thus increasing phosphorus and nitrogen removal via nitrite. Comparing the in-situ with the ex-situ experiments, notable differences are observed. In all cases higher rates were obtained for the in-situ experiments for both phosphorus and nitrogen. However, in the cases of liquid effluents from OFMSW the rates are relatively comparable. Only in the case of FL CM&MS the in-situ experiments resulted in much higher nitrogen and phosphorus removal rates than in the batch experiments.

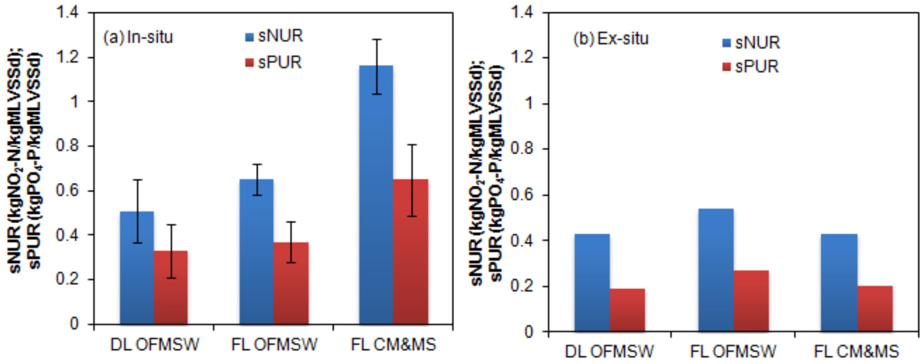

Figure 3: Effect of the external carbon source type on the nitrogen (sNUR) and phosphorous (sPUR) specific uptake rate under anoxic (NO_2-N) conditions in (a) in-situ and (b) ex-situ experiments

The cost analysis conducted by Frison et al. (2013) showed that FL OFMSW was the best available external carbon source, since the estimated cost per kg of COD added and of nitrogen removed were much lower (i.e. <0.10 €/kgCOD_{added} and 0.10 €/kg$N_{removed}$) than the respective costs of FL CM&MS (0.51 €/kgCOD_{added} and 0.74 €/kg$N_{removed}$). Figure 4 shows the sPUR/sNUR ratio for the three examined carbon sources. The phosphorus removed to nitrogen that is denitrified was always higher in the in-situ experiments reaching 0.63 kgPO_4-P_{uptake} /kgNO_2-N_{rem} for DL OMFSW and approximately 0.57 kgPO_4-P_{uptake} /kgNO_2-N_{rem} for the other two carbon sources.

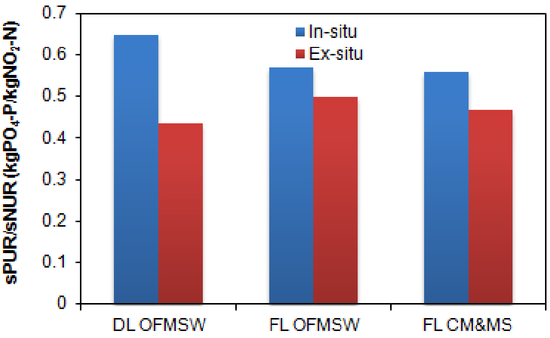

Figure 4: sPUR/sNUR ratio of the in-situ and ex-situ experiment

4. Conclusions

The three examined biowaste derived carbon sources resulted in significant denitrification (sNUR = 0.51-1.16 kgNO_2-N/kgVSSd) and phosphorus uptake (sPUR = 0.33-0.65 kgPO_4-P/kgVSSd) via the nitrite pathway. Therefore, the examined biowaste derived liquids can be used effectively for the via nitrite nutrients removal instead of conventional carbon sources which are commercially available.

Acknowledgments

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