

Use of external carbon sources derived from biowaste for short-cut nutrient removal from anaerobic effluents

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ABSTRACT

This work evaluated the use of different external carbon sources to promote the via-nitrite nutrient removal from anaerobic effluents. The carbon sources consisted of fermentation liquid produced from the organic fraction of municipal solid waste (OFMSW FL), drainage liquid produced from OFMSW, fermentation liquid produced from vegetable and fruit waste (VFW FL) and acetic acid. Denitrification and phosphorus uptake via nitrite were evaluated in two sequencing batch reactors, one treating the anaerobic supernatant produced from the co-digestion of OFMSW and activated sludge (highly nitrogenous anaerobic effluent – HNAE), and the other one treating the weakly nitrogenous anaerobic effluent (WNAE) from an upflow anaerobic sludge blanket reactor. The use of OFMSW FL to treat HNAE resulted in high nitrite (27 mgN/gVSS-h) (VSS – volatile suspended solids) and phosphate uptake (15 mgP/gVSS-h). In the WNAE, nutrient kinetics were much slower. The use of acetic acid and VFW FL performed poorly, while the use of OFMSW FL, which was rich in butyric acid and propionic acid, resulted in significant nutrient removal (7 mgN/gVSS-h and 6 mgP/gVSS-h). The economic evaluation showed that the use of OFMSW FL is a less expensive option than the acetic acid use.

Key words | anaerobic effluents, biowaste, external carbon source, nutrient removal via nitrite

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INTRODUCTION

Short-cut nitrogen removal through ammonium oxidation to nitrite and its subsequent reduction to gaseous nitrogen instead of conventional nitrification/denitrification has gained increasing attention over the last years due to its inherent advantages (Peng & Zhu 2006). The denitrifying via nitrite biological phosphorus removal (DNBPR) integrates phosphorus and nitrogen removal in a robust process in which the via-nitrite denitrification and phosphate uptake occur simultaneously. DNBPR can increase the economic and environmental sustainability of the process. It requires alternating anaerobic and anoxic conditions to promote the growth of the denitrifying phosphorus-accumulating organisms (DPAOs). The process requires significant concentration of short-chain fatty acids (SCFAs), which are taken up by DPAOs to form polyhydroxyalkanoates (PHAs). Apart from the SCFAs concentration, their composition has also been reported to affect the performance of the process (Feng *et al.* 2009). A key parameter is the distribution of the chain length of the volatile fatty acids (VFAs) (Lee *et al.* 2014). SCFAs, such as

propionic acid and butyric acid are the most suitable substrates to enhance DNBPR (Chen *et al.* 2004). Recent studies have shown that carbon sources containing a mixture of SCFAs can improve DNBPR (Ji & Chen 2010; Li *et al.* 2011). The addition of chemically synthesized SCFAs to improve biological nutrient removal increases the operating expenses, as the price of commonly used synthetic carbon sources has increased significantly over the last decade. The increasing oil price has renewed the interest in biological routes of SCFA production (Akaraonye *et al.* 2010; Lee *et al.* 2014). The research of the optimum and cost-effective carbon source to enhance nutrient removal is of growing concern (Gustavsson 2010). Significant cost savings can arise when the applied external carbon source is derived from biodegradable organic waste (e.g. fermented sewage sludge, food waste) instead of commercially available chemicals (i.e. acetic acid) (Mayer *et al.* 2009; Ji & Chen 2010). Following this approach, the amount of waste generated is reduced, through the conversion of waste into valuable products. Anaerobic effluents usually have a low

content of biodegradable organic matter; this reveals the need for external carbon source supply in order to accomplish effective nutrient removal. This is true both for highly nitrogenous anaerobic effluents (HNAE), such as the anaerobically digested pig slurry, leachate and reject water in municipal wastewater treatment plants (WWTPs), and for weakly nitrogenous anaerobic effluents (WNAE), such as anaerobically treated domestic and municipal wastewater. Ideally, the external carbon source should maximize the kinetics of nitrogen and phosphorus removal and have zero cost. The use of fermentation liquids as external short-chain carbon sources is the most common and effective practice. Several studies have examined the performance of short-cut nitrification/denitrification by applying sludge fermentation liquid in bench, pilot and demonstration scale systems (Mayer *et al.* 2009; Ji & Chen 2010; Zhang *et al.* 2010). Food waste can be used as a valuable source of VFA production (Elbeshbishy *et al.* 2011; Lee *et al.* 2014).

In this work, an economic and kinetics evaluation of different carbon sources used for the via-nitrite nutrient removal was carried out. Three biowaste-derived external carbon sources and acetic acid were tested to enhance nitrification/denitrification and DNBPR for the treatment of the anaerobic effluents. Furthermore, an economic analysis was carried out to demonstrate the sustainability of using biowaste-derived carbon sources for nutrient removal.

MATERIALS AND METHODS

Sequencing batch reactors treating anaerobic effluents

The via-nitrite nutrient removal process was investigated in two short-cut sequencing batch reactors (scSBRs). One scSBR was used to treat the anaerobic supernatant resulting from the co-digestion of waste activated sludge (WAS) and the organic fraction of municipal solid waste (OFMSW) and the other scSBR was used to treat the anaerobic effluent produced from the treatment of domestic wastewater by an upflow anaerobic sludge blanket (UASB). The former scSBR was located at the municipal WWTP of Treviso, in Veneto region (North Italy). The pilot scSBR had a working volume of 2.8 m³ and could treat up to 6 m³/d of anaerobic supernatant of WAS and OFMSW (Frison *et al.* 2012). The scSBR was equipped with two peristaltic pumps, which allowed the dosing of caustic soda (NaOH 30% wt) to adjust the pH and the supply of the external carbon source. The dosing of the external carbon source was

conducted during the first 1–2 min of the anoxic phase. The aeration was provided with the use of three blowers that were automatically switched on/off through the monitoring of the dissolved oxygen (DO) concentration in the reactor. A programmable logic controller (PLC, National Instruments – cRIO-9075) processed the analog signals that were recorded by the submerged probes. These probes included the pH (Hach Lange), the oxidation reduction potential (ORP, Chemitec and Hach Lange), the ammonium (NH₄D sc Hach Lange), the conductivity (Hach Lange), the NO_x-N (Nitratax plus Hach Lange coupled with the sample filtration system Filtrax Hach Lange), the mixed liquor suspended solids (Solitax Hach Lange) and the temperature (PT100). The scSBR operated at an average temperature of 17 ± 2 °C, while the system's heating was set in order to avoid temperatures lower than 15 °C. The hydraulic retention time (HRT) was 0.52 d and the sludge retention time was higher than 20 d.

The second scSBR was installed at the laboratory of the University of Verona and treated 50–90 L/d of UASB effluent. The UASB reactor treated synthetic domestic wastewater. The scSBR had a working volume of 26 L and was fed with low strength UASB effluent from two (100 L each) storage tanks using a suitable peristaltic pump. Online submerged probes (Hach Lange) of DO, pH, ORP, conductivity and temperature were used to monitor the process. The SBR operated at room temperature (22 ± 3 °C). The probe signals were processed and stored by a PLC (Compact Field Point CF-2200, National Instruments).

External carbon sources from biowaste

Four different types of external carbon source were tested: drainage liquid produced from the organic fraction of municipal solid waste (OFMSW DL), fermentation liquid produced from OFMSW (OFMSW FL) and from vegetable and fruit waste (VFW FL) through controlled acidogenic fermentation, and commercially available acetic acid (80% wt). The OFMSW DL was the leachate residual produced during the storage of raw OFMSW in the full scale storage pit of the Treviso WWTP. During the storage of OFMSW, the production and the accumulation of OFMSW DL was approximately 70–140 L/t of raw OFMSW. The accumulation depended on the temperature and the seasonal variations of the raw OFMSW. The OFMSW DL was produced from uncontrolled fermentation activities, due to the prolonged retention time in the storage pit (higher than 1–2 days). However, the term of DL is used in order to distinguish this type of carbon source from the OFMSW FL produced from controlled

fermentation. The OFMSW FL was produced through a sequencing batch fermentation unit receiving pre-sorted and ground OFMSW in a 200 L reactor. The source-separated OFMSW was ground and diluted with secondary effluent up to 6% total solids (TS); the organic loading rate was maintained at 20 kg total volatile solids (TVS)/(m³·d) with a HRT of 3 days (Pavan et al. 1998). The reaction temperature was maintained at 37 °C with the use of an electric water heater. The VFW FL was also produced through the controlled fermentation of pre-sorted vegetables and fruits using the operating conditions described above in a 30 L batch fermentation reactor. The main difference was the lower temperature of the process (26–28 °C).

Biomass activity tests

The via-nitrite denitrification and phosphorus uptake activity were determined through *in situ* experiments in the scSBRs. For this purpose samples were collected from the scSBRs every 10–30 min during the anoxic and aerobic phases. The samples were filtered and their NO₂-N, NO₃-N and PO₄-P concentrations were measured to determine the specific nitrite uptake rate (sNUR) and the specific phosphate uptake rate (sPUR) as well as the nitrogen and phosphorus removal rates (NRR and PRR respectively).

Analytical methods

TS, TVS, total suspended solids, volatile suspended solids (VSS), pH, chemical oxygen demand (COD), total Kjeldahl nitrogen (TKN), ammonium (NH₄-N), and phosphorus were determined by *Standard Methods* (APHA et al. 1998). Nitrite (NO₂-N), nitrate (NO₃-N), and phosphate (PO₄-P) were determined by ion chromatography (Dionex IC-90 with AG14 and AS14 columns). The composition of the SCFAs was determined by gas chromatography using a Nukol 15 m column, working at 85–125 °C (rate 30 °C/min) and nitrogen as carrier gas (flow 5 mL/min).

RESULTS AND DISCUSSION

Physicochemical characteristics

Table 1 shows the physicochemical characteristics of the HNAE and the WNAE. Although these effluent streams present very different characteristics, they both have a very low

Table 1 | Physicochemical characteristics of the HNAE and the WNAE fed to the SBRs

| Parameter | HNAE | WNAE |
|---------------------------|-------------|--------------|
| pH | 7.73 ± 0.23 | 7.85 ± 0.15 |
| Total COD (mg/L) | 108 ± 32.7 | 102.3 ± 42.2 |
| Soluble COD (mg/L) | 93 ± 27.4 | 67.5 ± 32.3 |
| TKN (mg/L) | 619 ± 86.7 | 59.7 ± 10.8 |
| NH ₄ -N (mg/L) | 586 ± 99.6 | 55.5 ± 17.3 |
| P (mg/L) | 95 ± 34.2 | 11.7 ± 2.2 |
| PO ₄ -P (mg/L) | 90.1 ± 24.3 | 9.5 ± 2.1 |
| COD/N (gCOD/gN) | 0.19 ± 0.05 | 1.74 ± 0.52 |
| Conductivity (ms/cm) | 4.21 ± 0.53 | 1.32 ± 0.11 |

COD/N ratio and require the use of external organic carbon source to achieve effective nutrient removal.

Table 2 shows the physicochemical characteristics of the various carbon sources, which were employed for the low and high strength nitrogenous effluents. The DL was characterized by significant VFA content, the majority of which was acetic acid. The significant VFA content shows that some uncontrolled fermentation had taken place due to the prolonged retention in the storage pit of OFMSW. The proportions of the different acids in the OFMSW DL and in the OFMSW FL were similar, showing that the controlled fermentation process did not alter the VFA proportions. However, the fermentation process increased the production of C2-C7 VFAs to 18.2 gCOD/L. On the other hand, the OFMSW DL contained only 6.9 gCOD/L of C2-C7 VFAs.

The OFMSW FL contained significant concentrations of butyric acid and propionic acid. On the other hand, the vast majority of VFAs (>90%) in VFW FL consisted of acetic acid, while other VFAs were contained in low concentrations. This is mainly related to the type of waste fermented, which consisted only of vegetable and fruit waste. Traverso et al. (2000) found that the acidogenic fermentation of VFW from supermarkets at mesophilic temperature resulted in the production of a liquid that was rich in acetic acid and lactic acid, but had very low content of butyric acid and propionic acid. Previous studies have shown that carbon sources containing a mixture of butyric acid and propionic acid can improve biological nutrient removal via nitrite (Ji & Chen 2010; Frison et al. 2013). Comparing the OFMSW FL that was dosed to treat the HNAE with the OFMSW FL dosed to the WNAE, the proportions of acetic acid, butyric acid and propionic acid were very different. In the second type of OFMSW FL, a much higher concentration of butyric acid and propionic acid was obtained after the fermentation process.

Table 2 | External carbon source characteristics

| Parameter | OFMSW DL HNAE | OFMSW FL HNAE | VFW FL WNAE | OFMSW FL WNAE |
|---------------------------|---------------|---------------|---------------|---------------|
| pH | 4.48 ± 0.09 | 4.14 ± 0.08 | 4.21 ± 0.22 | 4.25 ± 0.13 |
| TS (g/L) | 6.08 ± 0.47 | 25.37 ± 4.39 | 37.33 ± 8.59 | 44.21 ± 3.09 |
| TVS (g/L) | 5.03 ± 0.42 | 22.19 ± 2.43 | 24.87 ± 3.14 | 29.79 ± 2.13 |
| COD (g/L) | 57.79 ± 6.19 | 62.22 ± 5.37 | 34.39 ± 15.90 | 43.27 ± 73.51 |
| Soluble COD (g/L) | 41.81 ± 9.34 | 31.33 ± 3.09 | 27.76 ± 14.42 | 28.51 ± 10.42 |
| TKN (mg/L) | 2,207 ± 413 | 1,128 ± 319 | 730 ± 537 | 818 ± 198 |
| NH ₄ -N (mg/L) | 1,133 ± 31 | 315 ± 62 | 109 ± 68 | 268 ± 47 |
| P (mg/L) | 134 ± 22 | 540 ± 70 | 91.0 ± 47.6 | 292 ± 55.1 |
| PO ₄ -P (mg/L) | 93 ± 18 | 119 ± 42 | 55 ± 38 | 209 ± 35 |
| Acetic acid (mgCOD/L) | 4,400 ± 200 | 13,300 ± 700 | 3,178 ± 762 | 6,281 ± 923 |
| Butyric acid (mgCOD/L) | 800 ± 400 | 2,900 ± 400 | 193 ± 132 | 6,721 ± 872 |
| Propionic acid (mgCOD/L) | 500 ± 300 | 1,800 ± 300 | 7.2 ± 12 | 5,100 ± 592 |
| C5–C7 (mgCOD/L) | 1,200 ± 400 | 200 ± 100 | 50 ± 20 | 2,871 ± 434 |

Effect of carbon source on via-nitrite nutrient removal

Table 3 shows the via-nitrite nitrogen and phosphorus uptake (i.e. sNUR, sPUR_{anoxic}) and removal rates for the different types of external carbon sources that were dosed to treat low and high strength anaerobic effluents. Nitrogen removal rates were determined considering that nitrification was not limiting the process. As expected the kinetics were faster for the HNAE. Furthermore, the type of external carbon source that was fed to the scSBR had a significant impact on the removal rate of both nitrogen and phosphorus. Specifically, acetic acid resulted in a very high rate of the via-nitrite nitrogen removal from HNAE. The use of OFMSW FL and OFMSW DL resulted in high nutrient removal for the HNAE. OFMSW FL resulted in slightly higher removal of nutrients compared to OFMSW DL owing to the more controlled fermentation conditions. In the WNAE the use of OFMSW FL resulted in much higher via-nitrite nutrient removal compared to acetic acid and to VFW FL. This is attributed to the high concentrations of butyric acid and

propionic acid. The total COD/N ratio fed to the SBR was higher for the period when acetic acid was dosed compared to the respective ratio when VFW FL and OFMSW FL were added for the treatment of WNAE (Table 3). Despite this, the sNUR and sPUR were much lower in the case of acetic acid use compared to OFMSW FL.

In line with this work, the results from recent studies have shown that carbon sources that contain a mixture of SCFAs, such as butyric acid and propionic acid, can improve the DNBPR (Ji & Chen 2010; Li et al. 2011, Frison et al. 2013). Ji & Chen (2010) achieved 97.6% of soluble phosphorus removal from low strength effluents by applying the via-nitrite process using sludge fermentation liquid as an external carbon source, while the use of acetic acid resulted in 73.4% removal of soluble phosphorus. It has been demonstrated that propionic acid enhances the accumulation of PHA in sludge (up to 10–15% TVS), which allows the simultaneous denitrification and phosphorus removal in the presence of nitrite (Oehmen et al. 2005, 2006; Carvalho et al. 2007). The presence of propionic acid and butyric

Table 3 | Nitrite and phosphate via nitrite uptake, and removal rates for different external carbon sources in SBRs

| External carbon source | Treated effluent | COD/N (gCOD/gN) | sNUR (mgN/(gVSS · h)) | sPUR _{anoxic} (mgP/(gVSS · h)) | NR (kgN/(m ³ · d)) | PRR _{anoxic} (kgP/(m ³ · d)) |
|------------------------|------------------|-----------------|-----------------------|---|-------------------------------|--|
| Acetic acid | HNAE | 2.1 | 47.50 ± 5.28 | – | 1.09 ± 0.06 | – |
| OFMSW DL | HNAE | 2.7 | 21.25 ± 5.83 | 13.75 ± 5.00 | 0.49 ± 0.13 | 0.31 ± 0.11 |
| OFMSW FL | HNAE | 3.1 | 27.08 ± 2.92 | 15.42 ± 3.75 | 0.62 ± 0.07 | 0.35 ± 0.09 |
| Acetic acid | WNAE | 3.3 | 2.96 ± 1.37 | 0.62 ± 0.81 | 0.069 ± 0.032 | 0.008 ± 0.003 |
| VFW FL | WNAE | 3.2 | 3.11 ± 0.86 | 1.02 ± 0.87 | 0.067 ± 0.02 | 0.022 ± 0.007 |
| OFMSW FL | WNAE | 2.5 | 6.81 ± 1.99 | 6.33 ± 4.28 | 0.132 ± 0.04 | 0.122 ± 0.037 |

acid seems to favor more the DPAOs than the glycogen-accumulating organisms. When sludge fermentation liquid containing propionic acid and butyric acid is dosed, it results in a higher nitrite reductase activity of biomass compared to that obtained for acetic acid (Ji & Chen 2010).

Economic analysis of the applied carbon sources

The cost of the applied carbon source is critical, since it significantly affects the full scale application of the via nitrite process for nutrient removal from high and low strength anaerobic effluents. An economic analysis was carried out to determine the specific cost required for nitrogen removal via nitrite in €/kgN_{removed} using commercially available acetic acid and biowaste derived carbon sources. To conduct this analysis several parameters were taken into consideration; these include the specific consumption of the COD of the raw material per kg of nitrite removed, the deprived biogas (and thus energy) due to the diversion of the organic carbon source away from the anaerobic digestion process, the calorific value of methane (9200 kcal/m³CH₄) and the COD content of the fermentation liquid (1 gCOD/gTS), the cost of the raw materials, and the cost (or gate fee) for the treatment of the OFMSW (€75/t OFMSW). In terms of acetic acid, a cost of €700/t of 80% (wt) acetic acid was used (Frison et al. 2013), while for the waste-derived products no production cost was considered. The revenues from selling electrical energy produced from biogas were considered as €0.15/kWh. This is a realistic value, which considers the gradual reduction in the price that is taking place in several EU countries in which the specific cost used to be higher than €0.20/kWh. The cost of buying energy was taken as €0.10/kWh. In the analysis the more adverse option of using both the heating and electrical energy was considered. Consequently, the use of OFMSW as carbon source deprives energy both in terms of heat and electricity. The combined heat and power unit was considered to produce 35% electrical energy and 55% thermal energy from biogas. The economic analysis did not consider the cost of a base required to increase the pH of the carbon sources, as this is also required for commercially available SCFAs, nor the energy required to heat the fermenter, as this can be covered by the heat generated by the anaerobic digestion unit. Furthermore, the capital and operating expenses of the fermentation reactor were not considered in the analysis.

Table 4 shows the amount of methane that is lost when the waste-derived carbon source is diverted from the anaerobic treatment process and is used in the SBR for nutrient removal. This amount is relatively similar for the different carbon

Table 4 | Economic evaluation of external organic carbon source addition for the via-nitrite nitrogen removal (capital and operating expenses of the fermentation unit have been neglected)

| Parameter | Acetic acid | OFMSW DL HNAE | OFMSW FL HNAE | VFW FL WNAE | OFMSW FL WNAE |
|---|-------------------|---------------|---------------|-------------|---------------|
| Savings from OFMSW gate fee (€/kgN _{removed}) | – | – | 0.93 | 0.96 | 0.75 |
| Methane deprived (Nm ³ CH ₄ /kgN _{removed}) | – | – | 1.09 | 1.12 | 0.88 |
| Total energy deprived (kWh/kgN _{removed}) | – | – | 11.61 | 11.98 | 9.36 |
| Deprived revenue (€/kgN _{removed}) | – | – | 1.25 | 1.29 | 1.01 |
| Net specific cost of carbon source use (€/kgN _{removed}) | 1.72 ^a | – | 0.32 | 0.33 | 0.26 |

^aThe cost was calculated by considering the COD/N ratio of 2.1.

sources ranging from 0.88 to 1.12 Nm³CH₄/kgN_{removed}. Furthermore, the diversion of OFMSW away from the usual treatment process has a net income as the gate fee for the collection and treatment of OFMSW is not paid. The use of OFMSW FL results in much lower specific cost than acetic acid use, even when the revenues that are lost due to the diversion of OFMSW towards fermentation instead of anaerobic digestion are considered. However, this analysis does not consider the cost resulting from the operation and maintenance of the fermentation unit. Although the heat requirements could be covered by the heat generated by the anaerobic digestion process, energy is also required for the solid/liquid separation process. In the future, the use OFMSW FL is expected to be an even more attractive solution, as the cost of petrochemically derived products increases, while the tariffs for selling electricity from the production of biogas through OFMSW anaerobic digestion decrease. These features are useful to estimate the payback time for investments in fermentation plants for the *in situ* production of best available carbon source.

CONCLUSION

The OFMSW FL resulted in high via-nitrite nutrient removal from HNAE (27 mgN/gVSS·h and 15 mgP/gVSS·h) and significant removal from WNAE (7 mgN/gVSS·h and 6 mgP/gVSS·h). The VFW FL, which was primarily composed of acetic acid, resulted in much lower nutrient removal rates from WNAE than the OFMSW FL. Acetic acid resulted in high nitrogen removal rate from HNAE, but it performed poorly when it was added to WNAE. On

the basis of the economic evaluation, the OFMSW FL was the most beneficial option. Therefore, this cost can be considered to evaluate the payback time of fermentation plant for *in situ* production of best available external carbon.

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