Influence of liquid-to-biogas ratio and alkalinity on the biogas upgrading performance in a demo scale algal-bacterial photobioreactor

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**ABSTRACT**

The influence of the liquid-to-biogas ratio (L/G) and alkalinity on methane quality was evaluated in a 11.7 m\(^3\) outdoors horizontal semi-closed tubular photobioreactor interconnected to a 45-L absorption column (AC). CO\(_2\) concentrations in the upgraded methane ranged from <0.1 to 9.6% at L/G of 2.0 and 0.5, respectively, with maximum CH\(_4\) concentrations of 89.7% at a L/G of 1.0. Moreover, an enhanced CO\(_2\) removal (mediating a decrease in CO\(_2\) concentration from 9.6 to 1.2%) and therefore higher CH\(_4\) contents (increasing from 88.0 to 93.2%) were observed when increasing the alkalinity of the AC cultivation broth from 42 ± 1 mg L\(^{-1}\) to 996 ± 42 mg L\(^{-1}\). H\(_2\)S was completely removed regardless of the L/G or the alkalinity in AC. The continuous operation of the photobioreactor with optimized operating parameters resulted in contents of CO\(_2\) (<0.1%–1.4%), H\(_2\)S (<0.7 mg m\(^3\)) and CH\(_4\) (94.1%–98.8%) complying with international regulations for methane injection into natural gas grids.

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1. Introduction

The anaerobic digestion (AD) of organic solid waste and sludge from wastewater treatment generates a biogas that represents a potential renewable energy source capable of generating electricity and reduce the dependence on fossil fuels (Muñoz et al., 2015). Biogas can be purified and injected into natural gas grids or used as a vehicle fuel, or desulphurised and used for the generation of domestic heat or steam and electricity in industry (Andriani et al., 2014; Muñoz et al., 2015). In this regard, a growing contribution of biogas to the EU energy sector has been observed within the past years, with an increase in the numbers of biogas producing plants by a factor of 3 (from 6772 in 2009 to 17,439 by the end of 2016) (European Biogas Association, 2017). The upgrading of biogas prior injection into natural gas grids or use as a vehicle fuel is required due to the large number and high concentrations of impurities in raw biogas: CO\(_2\) (15–60%), H\(_2\)S (0.005–2%), O\(_2\) (0–1%), N\(_2\) (0–2%), CO (< 0.6%), NH\(_3\) (< 1%), siloxanes (0–0.2%) and volatile organic compounds (< 0.6%) (Ryckeboch et al., 2011). In this context, most international regulations establish that a methane composition of CH\(_4\) ≥ 95%, CO\(_2\) ≤ 2–4%, O\(_2\) ≤ 1% and negligible amounts of H\(_2\)S is mandatory for its injection into natural gas grids, while a lower CH\(_4\) content is required when methane is used as a vehicle fuel (Muñoz et al., 2015). The removal of biogas contaminants like H\(_2\)S reduces the corrosion in pipelines, engines and biogas storage structures, while the reduction in CO\(_2\) contributes to increase the calorific value of methane and reduces its transportation costs (Posadas et al., 2015).

Nowadays, several biological technologies are available to remove CO\(_2\) and H\(_2\)S from biogas. For instance, chemoautotrophic biogas upgrading is used for the removal of CO\(_2\), while biofiltration or in situ micro-aerobic AD are applied for H\(_2\)S removal (Farooq et al., 2018; Marín et al., 2018a; Muñoz et al., 2015). The removal of only one biogas contaminant at a time represents the main disadvantage associated to these biological technologies, resulting in the need of implementing two-stage biological upgrading processes. Likewise, several physical-chemical technologies are commercially available to remove CO\(_2\) and H\(_2\)S from biogas. Membrane separation, pressure swing adsorption or chemical/water/organic scrubbing are applied for CO\(_2\) removal.
removal, while in-situ chemical precipitation or adsorption onto activated carbon or metal ions provide satisfactory levels of H2S removal (Marín et al., 2018a; Muñoz et al., 2015; Toledo-cervantes et al., 2017). Two sequential stages are also necessary for a complete biogas upgrading, which entails an increase in investment and operational costs. In this context, algal-bacterial photobioreactors can be engineered as an environmentally friendly and cost-effective technology due to their capacity to simultaneously remove CO2 and H2S in a single stage process (Bahr et al., 2014).

Algal-bacterial processes have emerged as a cost-competitive technology capable of removing CO2 and H2S from biogas in a single stage at low environmental impacts (Bahr et al., 2014; Muñoz et al., 2015). Biogas upgrading in algal-bacterial photobioreactors is based on the simultaneous photosynthetic fixation of CO2 by microalgae and the oxidation of H2S to SO4^{2−} by sulfur oxidizing bacteria promoted by the high dissolved oxygen (DO) concentration present in the cultivation broth as a result of photosynthesis (Posadas et al., 2017, 2015; Toledo-Cervantes et al., 2016). Photosynthetic biogas upgrading has been recently evaluated indoors in high rate algal ponds (HRAPs) interconnected to a biogas absorption column (AC) under artificial illumination. Bahr et al. (2014) demonstrated for the first time the capability of microalgal-bacterial processes for the simultaneous removal of CO2 and H2S from biogas. Serejo et al. (2015) studied the influence of the liquid/biogas (L/G) ratio on the composition of the upgraded biogas. Posadas et al. (2016) optimized the biogas upgrading process in a HRAP using centrates as a source of nutrients under laboratory conditions, while Rodero et al. (2018) evaluated the influence of alkalinity and temperature on the photosynthetic biogas upgrading efficiency in an indoor HRAP. In addition, Posadas et al. (2017) evaluated the simultaneous biogas upgrading and centrerate treatment in a HRAP operated under outdoors conditions during summer, while Marín et al. (2018a,b) investigated the influence of the yearly variations of environmental conditions on the biogas upgrading performance. Nevertheless, and despite the satisfactory results obtained so far, new photobioreactor configurations should be tested in order to overcome design constraints associated to algal ponds such as their high footprint. In this sense, semi-closed or closed tubular photobioreactors have been proposed as a promising alternative to reduce land requirement, while offering higher photosynthetic efficiencies, enhanced biomass productivities and a superior CO2 mass transfer (Toledo-Cervantes et al., 2018).

This study investigated for the first time the biogas upgrading potential of an outdoors pilot-scale hybrid (semi-closed) horizontal tubular photobioreactor (PBR) interconnected to an external AC. The influence of the L/G ratio and the alkalinity of the cultivation medium in the AC on the quality of the upgraded biogas was assessed and optimized. In addition, the PBR-AC was operated continuously under optimized process parameters.

2. Materials and methods

2.1. Biogas

The biogas used in this experiment was obtained from the anaerobic digestion of microalgal biomass in a pilot anaerobic digester located at the Agròpolis experimental campus of the Universitat Politècnica de Catalunya-BarcelonaTech (Catalunya, Spain) (García et al., 2018; Uggetti et al., 2018). The average biogas composition was CO2 (13.7 ± 1.0%), H2S (0.1 ± 0.05%) and CH4 (86.2 ± 1.0%).

2.2. Experimental set-up

The experimental set-up was built outdoors at the Agròpolis experimental campus of the Universitat Politècnica de Catalunya-BarcelonaTech (41.29°N, 2.04°E). The horizontal hybrid (semi-closed) tubular photobioreactor (PBR) consisted of 2 lateral open tanks made of polypropylene (width = 1 m; length = 5 m; depth = 0.6 m) interconnected by 16 low density transparent polyethylene tubes (length = 47 m; diameter = 125 mm). The total working volume of the PBR was 11.7 m³. The cultivation broth was continuously circulated in each tank by a 6-blade paddlewheel with a rotational speed of 9–12 rpm, which resulted in a velocity of the cultivation broth inside the tubes of 0.20–0.25 m s^{-1}. This recirculation rate ensured a homogeneous distribution and mixing of the cultivation broth and a turbulent flow inside the tubes, avoiding biomass settling. The different height level between the two open tanks caused a gravity flow through 8 tubes from the deep side of one tank to the shallow side of the opposite one (Uggetti et al., 2018). The open tanks supported the release of the DO accumulated along the closed tubes and also provided a cooling effect via water evaporation, thus preventing the occurrence of the extremely high temperatures that would be reached in completely closed tubular PBRs. The PBR was interconnected to a separate 45 L bubble AC (internal diameter = 12 cm; height = 4 m) made of PVC and provided with a ring of seven metallic biogas diffusers of 2 µm pore size located at the bottom of the column (Fig. 1).

Fig. 1. Schematic diagram of the experimental set-up used for the continuous photosynthetic upgrading of biogas.
2.3. Operational conditions and experimental procedure

The PBR was inoculated at an initial concentration of 220 mg volatile suspended solids (VSS) L\(^{-1}\) with a microalgal consortium composed of *Chlorella vulgaris*, *Stigeoclonium tenue*, *Nitzschia closterium* and *Navicula amphora*, obtained from an outdoors HRAP located at the facilities of the Environmental Engineering and Microbiology Research Group (GEMMA) the Universitat Politècnica de Catalunya-BarcelonaTech (Gutiérrez et al., 2016). The PBR was operated as the third of a set of 3 identical PBRs interconnected in series and treating 2.3 m\(^3\)d\(^{-1}\) of agricultural wastewater with the following composition: total organic carbon (TOC) = 131 ± 80 mg L\(^{-1}\), inorganic carbon (IC) = 36 ± 10 mg L\(^{-1}\), total nitrogen (TN) = 15 ± 7 mg L\(^{-1}\) and total phosphorus (TP) = 0.9 ± 1.0 mg L\(^{-1}\). Three experimental series were conducted as described below:

### 2.3.1. Influence of the liquid-to-biogas ratio in the absorption column on the quality of the upgraded biogas

L/G ratios ranging from 0.5 to 5.0 were tested in order to optimize the quality of the upgraded biogas. Biogas was sparged into the AC at 100 L d\(^{-1}\) while the cultivation broth from the PBR was supplied in co-current mode at different flow rates in order to provide L/G ratios of 0.5, 1.0, 2.0, 3.0, 4.0 and 5.0. The duration of each L/G ratio condition was at least four times the hydraulic retention time (HRT) of the liquid in the AC (Table 1). The ambient and cultivation broth temperatures, the pH, dissolved TOC, IC, TN, N-NH\(_4^+\) and TP concentrations in the cultivation broth of the PBR, and the composition of the raw and upgraded biogas were analyzed in triplicate at the end of each operational condition.

### 2.3.2. Influence of the alkalinity in the cultivation broth on the quality of the upgraded biogas

In order to assess the impact of different alkalinitities of the cultivation broth in the AC on the upgrading efficiency, a carbonate solution (NaHCO\(_3\) and Na\(_2\)CO\(_3\)) with a concentration of 16,000 mg L\(^{-1}\) of IC was injected at the bottom of the AC in co-current mode (Fig. 1, dashed line). Biogas flowrate and L/G ratio were fixed at 100 L d\(^{-1}\) and 0.5, respectively. Carbonate solution flowrates of 0, 1, 2, 3 and 5 L d\(^{-1}\) (corresponding to an IC concentration in the cultivation broth of the AC of 42 ± 1; 311 ± 6; 634 ± 48; 996 ± 42 and 1557 ± 26 mg L\(^{-1}\), respectively) were tested in order to optimize the quality of the upgraded biogas. Each carbonate solution flowrate was maintained for at least four times the HRT of the liquid in the AC. The ambient and PBR cultivation broth temperatures, the pH, dissolved TOC, IC, TN, N-NH\(_4^+\) and TP concentrations in the cultivation broth of the PBR, and the composition of the raw and upgraded biogas were analyzed in triplicate at the end of each operational condition.

### 2.3.3. Continuous photosynthetic biogas upgrading operation

Biogas upgrading performance of the demo scale PBR was evaluated throughout 42 days under continuous operation. The optimum operating parameters previously identified were selected: biogas flowrate of 100 L d\(^{-1}\), L/G ratio of 0.5 and the supplementation of 2.0 L d\(^{-1}\) of carbonate solution to the AC. The ambient and cultivation broth temperatures, the pH, dissolved TOC, IC, TN, N-NH\(_4^+\) and TP concentrations in the cultivation broth of the PBR, and the composition of the raw and the upgraded biogas were analyzed in duplicate once per week.

### 2.4. Analytical procedures

The concentration of CH\(_4\), CO\(_2\), N\(_2\) and O\(_2\) in biogas and methane were determined using a gas chromatograph (GC) equipped with a thermal conductivity detector (Trace GC Thermo Finnigan with Hayesep packed column). Injector, detector and oven temperatures were maintained at 150, 250 and 35 °C, respectively, with helium as a carrier gas. The concentration of H\(_2\)S in the raw biogas was determined using Gastec colorimetric tubes, while its concentration in the upgraded methane was analyzed by a Dräger X-am 5000 electrochemical sensor (lower detection limit of 0.5 ppmv). Temperature and pH were measured in-situ by a pH-meter with temperature sensor (Mettler Toledo, USA). Dissolved TOC, IC and TN concentrations were determined using a C/N analyzer (21005, Analytikjena, Germany). The analysis of TP concentration was performed according to the Ascorbic Acid Method of Standard Methods (APHA, 2005), while N-NH\(_4^+\) concentration was measured by a colorimetric method according to Solozzano (1969). The determination of the concentration of total suspended solids (TSS) and VSS in the PBR was performed according to Standard Methods (APHA, 2005), and the temperature of the cultivation broth was periodically monitored with a temperature sensor (Campbell Scientific Inc., USA).

### 3. Results and discussion

#### 3.1. Influence of the liquid-to-biogas ratio in the absorption column on the quality of the upgraded biogas

The composition of the methane produced in the PBR-AC varied depending on the L/G ratio tested (Fig. 2). At a L/G ratio of 2.0, CO\(_2\) was not detected in the upgraded methane, thus achieving minimum concentrations < 0.1% according to the GC detection limit. On the contrary, a maximum concentration of 9.6 ± 0.1% was recorded at a L/G ratio of 0.5 (Fig. 2). These results were in accordance with Posadas et al. (2017), who recorded the highest concentration of CO\(_2\) in methane at the lowest L/G ratio (≈12.0% at a L/G ratio of 0.5). L/G ratios > 2.0 supported a significant decrease in the CO\(_2\) concentration of the upgraded biogas, which ranged from < 0.1 to 1.4% (corresponding to removal efficiencies (REs) between 90.4 and > 99.9%). On the other hand, H\(_2\)S was not detected in the upgraded methane regardless of the tested L/G ratio, its complete removal being attributed to the high aqueous solubility of this biogas contaminant. An efficient removal of

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**Table 1**

Operational parameters during the evaluation of the L/G ratio in the AC.

<table>
<thead>
<tr>
<th>L/G ratio</th>
<th>Liquid flowrate (L d(^{-1}))</th>
<th>Biogas flowrate (L d(^{-1}))</th>
<th>Biogas HRT (h)</th>
</tr>
</thead>
<tbody>
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<td>100</td>
<td>10.8</td>
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<tr>
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<td>100</td>
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<tr>
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<td>200</td>
<td>100</td>
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</tr>
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<td>300</td>
<td>100</td>
<td>1.8</td>
</tr>
<tr>
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<td>400</td>
<td>100</td>
<td>1.4</td>
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<tr>
<td>5.0</td>
<td>500</td>
<td>100</td>
<td>1.1</td>
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</tbody>
</table>

**Fig. 2.** Concentration of CO\(_2\) (■), N\(_2\) + O\(_2\) (●) and CH\(_4\) (○) in the upgraded biogas at different L/G ratios.
H₂S from raw biogas in algal-bacterial PBRs with a negligible impact of the L/G ratio has been consistently reported both in outdoors (Posadas et al. 2017) and indoors HRAPs (Serejo et al. 2015).

Unfortunately, the concentrations of N₂ and O₂ recorded in the upgraded biogas increased from 3.4% at a L/G ratio of 0.5 to 11.9% at a L/G ratio of 5.0 (Fig. 2), which clearly indicated that the stripping of these gases from the recirculating cultivation broth was promoted at higher liquid flowrates (Sovechles and Waters, 2015). These results were in accordance with Toledo-Cervantes et al. (2016), who reported N₂/O₂ concentrations between 2.5 and 37.0% at L/G ratios ranging from 0 to 40 in a closed tubular photobioreactor. Likewise, Posadas et al. (2017) also reported an increase in N₂ and O₂ concentration in the upgraded biogas from 1.4 to 18.3% when the L/G ratio increased from 0.5 to 5, respectively. Similarly, Rodero et al. (2019) found N₂/O₂ concentrations ranging from 6.6 and 11.4% at L/G ratios ranging from 1.2 to 3.5 in an outdoors HRAP.

Finally, a maximum concentration of CH₄ of 89.7% in the upgraded biogas was recorded at a L/G ratio = 1 (Fig. 2). Interestingly, although further increases in the L/G ratio resulted in lower CO₂ concentrations, they also mediated a higher desorption of N₂ and O₂, which negatively impacted the final concentration of CH₄ in the upgraded biogas.

3.2. Influence of the alkalinity in the cultivation broth on the quality of the upgraded biogas

The supplementation of a carbonate solution to the AC resulted in an improved quality of the final methane. In this context, average concentrations of CO₂ of 9.6 ± 0.2; 2.6 ± 0.2; 1.3 ± 0.0; 1.2 ± 0.0 and 1.1 ± 0.2% were recorded at IC concentrations in the AC cultivation broth of 42 ± 1; 311 ± 6; 634 ± 48; 996 ± 42 and 1557 ± 26 mg L⁻¹, respectively (Fig. 3). The increase in CO₂-REs resulting from the addition of alkalinity (from 24.0 ± 0.2% at 42 ± 1 mg ICL⁻¹ to 91.9 ± 0.2% at 1557 ± 26 mg ICL⁻¹) was associated to the concomitant increase of pH in the cultivation broth of the AC (from 6.5 ± 0.1 at 42 ± 1 mg ICL⁻¹ up to 9.3 ± 0.0 at 1557 ± 26 mg ICL⁻¹). The beneficial effect of alkalinity on CO₂ removal performance has been previously reported in literature. For instance, Rodero et al. (2018) reported CO₂-REs of 97.8 ± 0.8, 50.6 ± 3.0 and 41.5 ± 2.0% during the operation of an indoors HRAP interconnected to an AC using a feeding nutrient solution with an average IC concentration of 1500 mg L⁻¹, 500 mg L⁻¹ and 100 mg L⁻¹, respectively. On the other hand, the higher solubility of H₂S compared to that of CO₂ also mediated complete removals of this biogas contaminant regardless of the alkalinity of the AC cultivation broth. These results were in accordance with Franco-Morgado et al. (2017), who reported values of H₂S-REs of 99.5 ± 0.5% throughout the operation of an indoors HRAP interconnected to an AC using a highly carbonated medium at a pH of 9.5. Likewise, Rodero et al. (2018) observed H₂S-REs of 100.0 ± 0.0, 94.7 ± 1.9 and 80.3 ± 3.9% using a feeding nutrient solution with an average IC concentration of 1500 mg L⁻¹, 500 mg L⁻¹ and 100 mg L⁻¹, respectively.

The N₂ and O₂ concentration in the upgraded biogas increased from 2.4% at an IC concentration of 42 ± 1 mg L⁻¹ to 6.1% at 1557 ± 26 mg ICL⁻¹ (Fig. 3). This increase was attributed to the enhanced N₂ and O₂ stripped out from the recycling cultivation broth mediated by the increase in medium salinity (which ultimately decreased the solubility of these gases). Finally, the lowest concentration of CH₄ in the upgraded biogas (88.0%) was recorded at an IC concentration of 42 ± 1 mg L⁻¹, increasing up to a maximum concentration of 93.2% at 634 ± 48 mg L⁻¹ (Fig. 3). Interestingly, higher carbonate supplementation rates did not result in an additional increase in the CH₄ content. The increased CH₄ concentration at higher alkalinity loads was attributed to the limited desorption of N₂ and O₂ when operating at the optimum L/G ratio and the high absorption efficiency of CO₂ and H₂S due to the acidic nature of these gases. Similar results were obtained by Rodero et al. (2018), who reported CH₄ contents of 98.9 ± 0.2, 80.9 ± 0.8 and 75.9 ± 0.7% at average IC feed concentrations of 1500, 500 and 100 mg L⁻¹, respectively. Therefore, the results herein obtained confirmed the key role of alkalinity on the methane quality.

3.3. Continuous photosynthetic biogas upgrading operation

The optimum operating parameters (i.e. L/G ratio of 0.5 and supplementation of a 16,000 mg ICL⁻¹ solution to the AC at a flowrate of 2.01 L d⁻¹) identified in Sections 3.1 and 3.2 were selected to test the performance of the PBR during the continuous upgrading of raw biogas coupled with the treatment of the mixed wastewater.

3.3.1. Biogas upgrading

The composition of the methane obtained exhibited a rather constant value along the 42 days of operation (Fig. 4). CO₂ concentrations ranged between < 0.1% and 1.4%, corresponding to REs > 91.0% (Fig. 4). The previous optimization of key operating parameters such as the L/G ratio and the alkalinity in the cultivation broth of the AC supported these consistent CO₂ removals. Similarly, Marín et al. (2018a) reported values of CO₂ concentration in the upgraded biogas ranging from 0.7 to 1.9% throughout the operation of an outdoors HRAP interconnected to an external AC. It is important to highlight that the CO₂ concentrations here obtained fulfilled most international regulations for methane, which require CO₂ concentrations ≤2–4% to be acceptable for injection into natural gas grids (Muñoz et al., 2015).
varied throughout the process with values ranging from 69.9 to 277.3 mg L\(^{-1}\) in the influent and from 90.4 to 217.0 mg L\(^{-1}\) in the effluent (Fig. 5). The low TOC-REs recorded were attributed to the low biodegradability of the mixture of agricultural and domestic wastewater used as influent to the PBR. Moreover, the significant water evaporation rates from the cultivation broth in the open tanks and the lysis of the microalgae generated during photosynthetic CO\(_2\) fixation likely contributed to increase the TOC concentration in the effluent in comparison to that of the influent, thus resulting in the negative TOC-REs observed. On the other hand, the dissolved IC concentration in the influent varied from 21.6 to 46.3 mg L\(^{-1}\) and from 29.8 to 91.8 mg L\(^{-1}\) in the effluent (Fig. 5). Although no correlation between the IC concentration in the effluent of the PBR and the addition of the carbonate solution in the AC was found due to the high dilution effect associated to the large volume and short hydraulic retention time of the PBR, the high values of pH in the PBR ranging between 7.9 and 8.9 might have promoted the increase in the IC concentration of the effluent supported by biogas absorption. Finally, no effective TN removal was observed during the entire experimental period, with dissolved TN concentrations in the influent (ranging from 9.1 to 25.0 mg L\(^{-1}\)) comparable to those recorded in the effluent (ranging from 11.1 to 25.9 mg L\(^{-1}\)) (Fig. 5).

4. Conclusions

This work constitutes, to the best of our knowledge, the first validation of photosynthetic biogas upgrading in a pilot-scale semi-closed PBR interconnected to an AC under outdoors conditions. Both the L/G ratio and the alkalinity in the AC were identified as key parameters influencing the quality of the final methane, with optimum values of 0.5 and 634 ± 48 mg L\(^{-1}\), respectively. The implementation of the optimum operating parameters during continuous operation resulted in a methane with CO\(_2\) concentrations of < 0.1%–1.4%, H\(_2\)S < 0.5 ppm, and CH\(_4\) contents of 94.1–98.9%, which complied with most international regulations for methane injection into natural gas grids.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.biortech.2019.02.029.

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