

Above ground part of common reed to enhance anaerobic co-digestion of farm biomasses: Potential, monitoring and efficiency

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1 *Above ground part of common reed to enhance anaerobic co-digestion of farm*  
2 *biomasses: potential, monitoring and efficiency.*

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6 Abstract

7 This paper shows that common reeds used in phytoremediation plants can  
8 successfully disposed of in anaerobic digestion reactors. At mesophilic condition the  
9 following were investigated: biogas yield resulting from an anaerobic co-digestion campaign  
10 of mixtures of cattle slurry, cheese whey and aboveground biomass of *Phragmites australis*,  
11 rates of production of methane, removal efficiencies of chemical oxygen demand (COD) and  
12 bio-methane yield (BMY). The resulting concentration of methane in the biogas was between  
13 53-56%. Maximum removal efficiencies for COD was 70%. The mixture of 50% cattle

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14 slurry, 40% cheese whey and 10% of aboveground parts of *Phragmites australis* achieved an  
15 Organic Loading Rate (OLR) of 3.3 g-VS/l-d with a methane yield of 241 l-CH<sub>4</sub>/kg-VS. A  
16 high digestate methane potential with a high OLR in the feed was also tested, with a result  
17 of 219 l-CH<sub>4</sub>/kg-VS.

18 **Keywords** Anaerobic co-digestion - Digestate methane yield - Whey - *Phragmites australis*  
19 – COD reduction

20

## 21 1. Introduction

22 EEC Directive 91/271, concerning urban and industrial wastewater, rules that the  
23 wastewater produced from farming and industrial production must now be subjected to  
24 treatment. A low cost solution are phytoremediation systems that are becoming widely used  
25 for the treatment of wastewater. The vegetal species used in most of these systems are part  
26 of the common reed family. The common reed is currently the most frequently used plant  
27 inside phytoremediation systems. They are also one of the most widespread vascular plants  
28 on Earth and are one of the dominant plants in Europe. *Phragmites australis* is a macrophyte  
29 often used to phytoremediate wastewater coming from urban sewage, but more frequently,  
30 from farming and its aboveground parts are generally cut twice a year.

31 To date, research has focused mainly on the strategies used in the big scale management  
32 of these plants (Hanssons et al, 2004, Kuhlman et al., 2013 and Risén et al., 2013). Showed

33 that the aboveground parts of the common reed can be used to produce bio-methane.  
34 However, very little research has been carried out with regard to anaerobic co-digestion  
35 (A.D.). A productive final destination for the aboveground parts of common reed have been  
36 studied by Risén et al. (2013). This allows mixing different kinds of biomasses that need to  
37 be treated before disposal. The co-digestion of different kinds of biomass brings the  
38 Carbon:Nitrogen ratio into the optimal range, indicated in 20/1 – 30/1 by Parkin and Owen  
39 (1986), 10/1 – 30/1 by Schattauer and Weiland (2004), but also enhances the biomethane  
40 yield.

41 This paper reports on a study of enhancement of common reed co-digestion. C:N ratio high  
42 value influences CH<sub>4</sub> production as the carbon cannot optimally be converted to CH<sub>4</sub>. The  
43 low values obtained for C:N ratio implies that the feedstock could result in a high total  
44 ammonia nitrogen (TAN) release and a high volatile fatty acids (VFAs) accumulation inside  
45 the digester (e.g. cattle slurry and whey co-digestion). As demonstrated by Parkin and Owen  
46 (1986) both TAN and VFAs are fundamental intermediates and potential inhibitors in the  
47 anaerobic digestion process. A working method to avoid excessive ammonia accumulation  
48 is to adjust low feedstock C:N ratios by adding high carbon content materials such as the  
49 aboveground biomass of *Phragmites australis*. Several examples of anaerobic co-digestion  
50 of different substrates can be found in scientific literature. Sosnowaki et al. (2003) studied

51 the co-digestion of sewage sludge and municipal solid waste (MSW). Callaghan et al. (2002)  
52 investigated co-digestion of cattle manure/slurry with different kinds of fruit, vegetable  
53 wastes and chicken manures. In both cases successful blending of high C:N and low C:N  
54 feedstock improve digester performance and bring the following benefits of co-digestion:  
55 dilution of potentially toxic ammonia, allowing for an increased loading rate and an improved  
56 biogas yield.

57 The present study investigated the feasibility of anaerobic co-digestion of a previously  
58 tested mixture (cow manure and whey) with the addition of aboveground parts of *Phragmites*  
59 *australis* let out from a phytoremediation plant that treats cheese factory wastewater. The  
60 purpose was to investigate how an important variation in the feeding mixture can affect the  
61 methane production during co-digestion. The quality criteria for anaerobic digestion of  
62 selected substrates were established, and the suitability of *Phragmites australis* in the co-  
63 digestion and achievable methane yields was determined. Furthermore, the digestate methane  
64 yield potential was evaluated, as well as the Chemical Oxygen Demand (COD) removal  
65 efficiencies, and energy assessments were performed.

66 A full scale AD process produces unstable digestate and considerable biogas can be  
67 obtained. An investigation was therefore performed to compare this residual biogas potential  
68 (BMP) with the measurement of the initial biogas potential (BMP of the feeding materials).

69 This investigation helped to evaluate the full-scale AD process. An experimental campaign  
70 was conducted using a Continuous Stirred Tank Reactor of 128 L under standard temperature  
71 and pressure conditions (STP). Two benefits were obtained: first, the *Phragmites australis*  
72 used for the replacement of the cheese whey was derived from a phytoremediation plant  
73 treating its biomass. Second, an addition of carbon inside the substrate brought a more  
74 optimal C:N ratio.

75

## 76 2. Materials and methods

77 A pilot scale anaerobic digester was used for conducting the test. It had the following  
78 principal components: anaerobic reactor, gasometer and feed chamber. The size of the reactor  
79 was not relevant in the evaluation of the methane yield as this parameter was calculated under  
80 Standard Temperature and Pressure (STP) and expressed as CH<sub>4</sub> liter/kilograms of volatile  
81 solids. The only related variable was the quantity of volatile solids inserted inside the reactor.

82

### 83 2.1 Pilot device

84 The reactor covered a surface of 1.20 m<sup>2</sup>, was 2.30 m height (Fig. 1). The reactor had a 316  
85 stainless steel tank realized by a cylinder 90 cm high with a diameter of 40.3 cm, closed by  
86 two caps on the top and on the bottom, for a total volume of 128 l and a working volume of

87 about 100 l. It was equipped by a mixer system with a blade propeller and a scraper on the  
88 bottom, both 316 stainless steel made and activated by a variable speed electric engine  
89 controlled by an inverter. The digester and the gasometer were equipped with a complete  
90 probe monitoring system including: a temperature probe inserted on one side of the reactor;  
91 a temperature and a pressure probes on the gas holder; a pH probe inserted inside the digester.  
92 The temperature was automatically controlled to remain inside mesophilic range and it was  
93 regulated by an electrical resistance (15 m). It was wrapped around the reactor and covered  
94 with insulating film to maintain the temperature near 35°C. The system was equipped with a  
95 small tank to collect condense, designed to be emptied automatically. The indirect measure  
96 of biogas yield was obtained with the movement of a slide-wired potentiometer, which was  
97 linked from one side with the gasometer upper parts and fixed with the chassis from the other.  
98 The operational pressure was about 9-10 mBar. The outlet pipe was equipped with a solenoid  
99 valve activated by a relay to allow the automatic discharge of the produced biogas. This  
100 system was described in details and used in a previous experience (Comino et al., 2012).

101

## 102 2.2 Phytoremediation plant

103 The constructed wetland for the dairy “Laiterie Cooperative Valdigne” in Morgex (Valle  
104 d’Aosta, Italy) was built both to respect the environmental resources and to give a contribute

105 to the development of such technology. It consists on a fat-removal unit and a basin for the  
106 storage and the distribution of the wastewater which precede three pythoremediation beds  
107 (Fig. 2): the first two are parallel and they work as submerged vertical flow wetland with  
108 gravel medium for a total area of 180 m<sup>2</sup>, the last is a submerged horizontal flow wetland  
109 with sand medium and a total area of 360 m<sup>2</sup>. These beds are planted with *Phragmites*  
110 *Australis* (CAV.) Trin. Ex Steud., with a density of about 4 plants/m<sup>2</sup>. At the time of the  
111 biomass harvest, the system is two years old. Deep description, system efficiencies and  
112 inlet/outlet wastewater analysis of the constructed wetland were already presented in a  
113 dedicated work (Comino et al., 2011).

114

### 115 2.3 Feed material

116 The feed biomass used for the realization of the two campaigns was composed of cow  
117 manure, cheese whey and the aboveground parts of *Phragmites australis*. The cow manure  
118 and the fresh cheese whey were both collected at the livestock farm “Fontanacervo” located  
119 in Villastellone (Turin – Italy). The collected biomass that was not immediately used to fill  
120 the reactors for the startup phase, was stored inside a refrigerator at about 4°C. The  
121 aboveground parts of *Phragmites australis* (almost 7 kg of fresh biomass) was collected from  
122 the phytoremediation plant described above and stored inside a 50 L barrel. The *Phragmites*



123 biomass was first spread on a large surface and dried on a thin layer for 24 h at about 60°C.  
124 Then it was chopped into a smaller size of about 2 mm (Mshandete et al. 2006). The volatile  
125 solids were equal to 9.1 % in the cow manure, 4.5% in the cheese whey and 81% in the dried  
126 *Phragmites*; BOD<sub>5</sub> values were obtained for cow manure and cheese whey and were equal to  
127 39,000 mg/l, 59,000 mg/l, the COD were equal to 120,000 mg/l, 74,400 mg/l respectively.  
128 The influent and effluent details are presented in Table 1 and Table 2.

129

#### 130 2.4 Startup tests

131 The reactor was initially filled with only cattle slurry to obtain a stable anaerobic digestion  
132 under batch feeding conditions. About 80 kg were used for the scaled-up device. During the  
133 startup phase it is important to fill in with only cattle slurry the reactor to avoid the risk of a  
134 process collapse due to the whey trend to acidify very rapidly (Comino et al. 2012). Startup  
135 tests lasted 42 days. The substrate was stirred every 2 days for 30 min and the produced  
136 biogas was analyzed to monitor the quality.

137

#### 138 2.5 Co-digestion test

139 After the startup phase co-digestion of test mixture was started. The feeding ratio was  
140 implemented as follow: the total processed quantity was equal to 55 kg of mixture (50%

141 cattle slurry, 40% cheese whey and 10% *Phragmites australis*). The slight differences  
142 between the startup quantity (80 Kg) and the co-digestion one (55 Kg) was due to facilitate  
143 the loading/unloading operations. So the first step was to reduce the total volume inside the  
144 anaerobic reactor from 80 to 55 Kg. Then the co-digestion feeding strategy was adopted and  
145 about 5 kg of substrate was removed and a 5 kg mixture (2.5 kg of manure, 2 kg of cheese  
146 whey and 0.5 kg of *Phragmites australis*) was loaded inside the reactor. Each test lasted 34  
147 days of fed-batch feeding, plus a week of anaerobic rest with no feeding (batch condition).  
148 Feeding of the reactor was done 3 times a week until the complete replacement of the initial  
149 biomass. Such as was observed in past experience no purging with nitrogen was necessary.  
150 It was observed that less than 1% oxygen in the reactor volume did not adversely affect the  
151 test. The probes inside the system were calibrated before the beginning of the test. The gas  
152 production was checked at least twice a day via remote control. For the campaign the  
153 demonstrated different mixing intensity effect (Kaparaju et al. 2008), and the consequence  
154 of particle size on biogas yield (Mshandete et al. 2006) were taken in consideration. The  
155 substrate was stirred every time a feeding operation was performed for about 30-45 min. The  
156 temperature inside the reactor was always maintained at  $35.5\text{ }^{\circ}\text{C} \pm 0.5$ . Between one feed and  
157 the other no stirring was performed inside the reactor. The gasometer was equipped with  
158 electronic controlled electro valves that allowed biogas discharge when reached pre-

159 established values. pH, temperatures, pressure and gasometer vertical movements were all  
160 monitored at 5-min intervals. After the last feed, the system was left undisturbed for almost  
161 7 days to evaluate biogas production stabilization. Substrate samples for the chemical  
162 analysis were collected at the end of the test (Tab. 2).

163

#### 164 2.6 Digestate methane yield tests (DMY)

165 After the co-digestion was started a monitoring test without discharge of any biomass from  
166 the previous one. Indeed it was expected that the digestate still contain a considerable amount  
167 of undigested organic matter with a high OLR and a relatively short HRT. The substrates  
168 were stirred every two days for 45-min when biogas samples were collected. Main systems  
169 parameters were constantly checked including methane concentration inside the biogas. The  
170 anaerobic reactor remained sealed during the experimentation period. After 32 days of  
171 retention, the test was concluded and samples collected for chemical analysis (Tab. 2).

172

#### 173 2.7 Bio-methane yield

174 To allow direct comparison with full-scale plants the bio-methane yield ( $BMY_1$ ) of the co-  
175 digestion test was determinate, by using the following equation presented by Schievano et al.  
176 (2011):

177 
$$BMY_1(\%) = \frac{BMP_{in} * TS_{in} - BMP_{out} * TS_{out}}{BMP_{in} * TS_{in}} * 100; \quad (1)$$

178

179 Where  $BMP_{in}$  is the bio-methane potential in the fed mixture,  $BMP_{out}$  is the bio-methane  
180 potential in the output digestate,  $TS_{in}$  are the total solids fed during the observed period and  
181  $TS_{out}$  are the total solids output with digestate during observed period. The  $BMY_1$  obtained  
182 with Eq. (1), can be compared to the effective specific methane produced (SMP) in a full-  
183 scale plants and calculated with the following equation:

184 
$$BMY_2(\%) = \frac{SMP}{BMP_{in}} * 100; \quad (2)$$

185

## 186 2.7 Chemical analysis and procedures

187 The feed materials and obtained digestate were stored at 4 °C immediately after sampling.  
188 Chemical analyses were performed within 48 h in an accredited laboratory.  $BOD_5$  was  
189 analyzed with the IRSA-CNR n°. 5100 A/94 method; COD with the IRSA-CNR n°. 5110/94  
190 method; pH samples with IRSA-CNR Quad 100 method 2080/94 and inside the reactors with  
191 pH probes. Density was evaluated with the EMRO/012/1999 method; 105 °C residual, and  
192 the 550°C residual as the Total Volatile Solids were obtained with the IRSA-CNR Quad. 64  
193 n°. 2.4.2/84 method. Ammonia was evaluated following the IRSA/APAT guidelines 29/2003  
194 method n°. 4030C. Volatile Fatty Acids (C1-C6) were measured with the EMGC 003/1999

195 method. The *Phragmites australis* calorific value was calculated according to the ASTM D  
196 240 method. A GA-2000 gas analyzer was used for the real time biogas monitoring. The  
197 monitored parameters were CH<sub>4</sub>, CO<sub>2</sub>, O<sub>2</sub>, CO and H<sub>2</sub>S, as well barometric pressure and  
198 relative pressure inside the gasometer. Organic loading rate (OLR) and hydraulic retention  
199 time (HRT) were calculated based on the regular additions. The data collected by the data  
200 logger were analyzed at the end of each test and processed using an own made database to  
201 evaluate different analytic parameters.

202

### 203 3 Results and discussion

#### 204 3.1 Startup test

205 During the first 42 days, limited biogas production can be observed (Fig. 3). The pH value,  
206 which is not shown, increased in the first 2 days from about 6.9 to 7.8. This behavior  
207 anticipated an increase in biogas production which then reached a maximum between days 4  
208 and 6 and then stabilized (Fig. 4a). A total of about 979 l of biogas was produced (Fig. 3).  
209 Considering that CH<sub>4</sub> inside the biogas was 55.4% the total amount of produced methane was  
210 543 l. Following the methodology to obtain the specific bio-methane productions on VS basis  
211 with a total of 3.088 kg of VS inserted in the reactor was obtained a 174.8 l-CH<sub>4</sub>/kg-VS for  
212 42 days of active anaerobic digestion. The startup followed the expected behavior and trend

213 of biogas production from cattle manure. Similar values were found by Amon et al. (2007),  
214 Brachtl (2000), Thomè-Kozmiensky (1995) and Comino (2010, 2012). The ranges for the  
215 production of biogas, cited above, are all similar to what was obtained during the startup,  
216 which was 317.03 l-biogas/kg-VS.

217

### 218 3.1 Co-digestion tests

219 The use of a biomass mixture with 50% cheese whey and 50% cattle manure was proved  
220 suitable and effective to produce methane (Comino et al. 2012). The addition of aboveground  
221 biomass of *Phragmites australis*, to this mixture, allows to evaluate if a reduction of whey  
222 could change biogas production in quality and quantity. The different composition inside the  
223 substrates shows the methane yield at increasing OLR.

224 After a start-up phase with an OLR established inside the optimal range at about 0.9 g-  
225 VS/l-d, the test investigated the scaled-up reactor behavior using an OLR values equal to  
226 3.28 g-VS/l-d (Tab. 3). The pH monitoring showed very smooth fluctuation during the test,  
227 the periodic unloading/loading operations were not clearly visible. The biogas production is  
228 presented in Figure 3 and the daily biogas production in Figure 4b. The final rest period of  
229 about a week showed a very smooth and progressive reduction of biogas production rate. No  
230 accumulation of undegraded material was observed inside the digester. The methane

231 proportion inside the produced biogas with the scaled-up reactor (50% cattle slurry, 40%  
232 whey and 10% *Phragmites*) had an average value of 54.1%, a maximum value of 58.9% and  
233 a minimum of 46.5% (Fig. 3). It started to stabilize at the end of the test, while during the  
234 most part of the experiment followed a performance related to the feed operation. The  
235 obtained methane yield was equal to 241 l-CH<sub>4</sub>/kg-VS with an OLR of 3.28 g-VS/l-d. In  
236 terms of COD were found the following values equal to 110 g/l, OLR= 137.5 g, COD/l<sub>R</sub> and  
237 HRT=44 days. As visible in Fig. 3 the reactor continued to produce biogas smoothly, the  
238 total produced volume was equal to 3534.8 l with a COD reduction of 53% (Tab. 5).

239

### 240 3.3 Digestate Methane Yield (DMY) tests

241 To confirm the data obtained during the campaign was taken in consideration the  
242 realization of a theoretical methane yield, but it will bring a series of uncertainties that will  
243 be very difficult to discuss. The model originally developed by Buswell and Boruff (1932),  
244 that is able to estimates theoretical methane concentration starting from the chemical  
245 composition of organic substrate (C, H, N and S), do not integrate the influence of lignin and  
246 assume a total transformation of the element in CH<sub>4</sub> (that is not true under real conditions).  
247 Also, carbon content of a feed material can be used in combination with Buswell's equation  
248 to estimate methane production, but it is necessary to assume what proportion of the feed

249 material is degraded in the process and in this case, with a mix of three different materials, it  
250 will be very difficult to evaluate a real value. Another issue was the fact that a good model  
251 needs a detailed database as input factors, and this was not the case as the used materials and  
252 mix ratios were used for the first time. For all these reasons it was preferred to realize a  
253 Digestate Methane Yield (DMY) test after the experiment, in this way was possible to  
254 estimate the residual biogas/methane potential. A tests with a batch-fed regime was  
255 performed, as described in paragraph 2.5, after the co-digestion one.

256 The test was carried out after the co-digestion with 50:40:10 mixture, an estimated OLR of  
257 about 1.1 g-VS/l-d, that produced a total quantity of biogas equal to 782.7 l. Considering a  
258 CH<sub>4</sub> proportion of 53.6%, this amount corresponded to 419,8 l of methane (Fig. 3). The  
259 digestate methane yield was 218.9 l-CH<sub>4</sub>/kg-VS with a VS estimated value of 1.9 kg,  
260 obtained from the chemical analysis taken at the end of the co-digestion test. This value  
261 indicates that the digestate potential is very high, but, as visible in Figure 4c, the daily  
262 production is steady. In a real scale process digestate can yield an important amount of biogas  
263 that could be used to produce electricity (i.e. 0.9 kWh per t/d with the above values). Data in  
264 literature shown that the methane yields obtained by several studies are sensible lower than  
265 the one presented in this paper (Hansen et al. 2006; Lethomaki et al. 2008; Menardo et al.  
266 2011).



267

### 268 3.4 Discussion

269 No direct comparison can be made with other studies, as this substrate was never tested  
270 before, but few experiences were conducted in the past about the digestion of common reed  
271 alone or in co-digestion with other organic substrate. Renborg (1984) and Brodin et al. (1988)  
272 reported a methane production of 180 l-CH<sub>4</sub>/kg-VS conducting a small-scale studies using  
273 summer harvested reed. Risén et al. (2013) realized a test using two 30L CSTR digesters  
274 with an operating temperature of 52°C and 24 days of HRT. The substrate was a mixture of  
275 common reed and five different fractions of organic material (Slaughterhouse waste – 38%;  
276 Cattle manure slurry – 30%; Milk plasma permeate – 15%; Sludge – 12%; Potato residues –  
277 5%) obtaining a methane production from reed addition of about 219 l-CH<sub>4</sub>/kg-VS. In a test  
278 conducted by the authors with pre-treated crop silage and cattle slurry the obtained value was  
279 equal to 249 l-CH<sub>4</sub>/kg-VS with an OLR of 5.15 g-VS/l-d (Comino et al. 2010). Another  
280 experiment conducted with 50% of cattle slurry and 50% of whey had shown a methane yield  
281 of 343.4 l-CH<sub>4</sub>/kg-VS with an OLR of 2.65 g-VS/l-d (Comino et al. 2012). Few experiments  
282 were carried out on co-digestion of cattle slurry and cheese whey. In the past, for these kind  
283 of substrates, anaerobic digestion was principally used as a wastewater treatment method,  
284 and not considered as a system to produce energy. Lehtomaki et al. (2007) in a study with

285 several energy crops and cow manure mixture, found methane yields between 149 and 268  
286 l-CH<sub>4</sub>/kg-VS. In the high OLR test conducted by Lindorfer et al. (2008) was found methane  
287 yields between 360 and 400 l-CH<sub>4</sub>/kg-VS. A mix of whey and cow manure with a 2:1 ratio  
288 was tested successfully by Lo and Liao (1989), they obtained a methane yield equal to 222  
289 l-CH<sub>4</sub>/kg-VS. The presented results show that anaerobic digestion of cattle slurry, cheese  
290 whey with *Phragmites australis* could be successfully achieved without the use of chemicals  
291 (for pH correction) with a 50:40:10 substrate. As the aboveground part of the *Phragmites*  
292 was dry (Table 1) a liquid fraction was required to maintain sufficiently wet the substrate.  
293 Normally to compensate this situation the substrate was diluted with water. However, this  
294 experiment demonstrated that whey can be used as an ideal substitute for water, as even if  
295 contribute to increase the overall COD, the COD reduction in both test campaign were greater  
296 than 50%. Whey also contribute to add needed nutrients and vitamins to the microbiota.

297 Other studies with different types of biomass mixture, Stewart (1980) and Jarvis et al.  
298 (1997), have observed a maximum OLR of 6.7 kg-TS/m<sup>3</sup>-d. Nordberg et al. (2007) have  
299 reached a maximum OLR equal to 3 g-VS/l-d in a trial with alfalfa silage that brought to a  
300 process breakdown. In past tests a working and productive process with an OLR value equal  
301 to 5.15 g-VS/l-d was obtained, but when reached a OLR of 7.78 g-VS/l-d the system rapidly  
302 breakdown (Comino et al. 2010). With a high OLR of 6.35 g-VS/l-d the mostly affected

303 parameter is the residual methane potential that, as described in the next paragraph, could  
304 still remain high.

305 The  $BMY_1$  indicated relatively high efficiency of the lab-scale digester (78%) (Table 3).  
306 Observing the daily biogas production (Fig.4b) can be noticed a couple of slight inhibition  
307 of the methanogenic activity, occurred between days 10 to 14 and 21 to 24, mainly due to a  
308 probably increased lignin content. Johnsson (1986) reported that is important that the reed is  
309 not harvested too late in the growing season, since the lignin content will be higher.

310 With the presented result, considering the test conducted with the scaled-up reactor it  
311 would be possible to obtain electricity production of about 2.9 kWh per 1 t/d with a CHP  
312 technology with a considered efficiency of 36%. This kind of mixture has an energy potential  
313 with the same order of magnitude of the energy crop and livestock waste co-digestion. The  
314 obtained value is lower if compared to what is possible to obtain with energy crops co-  
315 digestion, but the environmental benefits are much higher. Also the use of phytoremediation  
316 plants for anaerobic co-digestion is a new possibility that is still not investigated adequately.  
317 Different types of species, specifically used for this scope, more optimized mixture of  
318 biomass inside the reactor can bring higher electricity production.

319

320

321 4 Conclusion

322 The results of this study show that the co-digestion, without induced pH correction, of  
323 typical cheese factory biomasses and a small fraction of *Phragmites australis* coming from  
324 a phytoremediation plant, is efficient and effective. A mixture of 50% cattle slurry, 40%  
325 cheese whey and 10% of pretreated *Phragmites australis* achieved an OLR of 3.28 g-VS/l-d  
326 with a methane yield of 241 l-CH<sub>4</sub>/kg-VS. The digestate methane potential with a high OLR  
327 in the feed was found equal to 219 l-CH<sub>4</sub>/kg-VS.

328

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409

Parameter	Cow manure	Cheese Whey	<i>Phragmites</i>
pH	6.94	4.12	-
BOD 5 (mgO <sub>2</sub> /l)	39,000	59,000	-
COD (mgO <sub>2</sub> /l)	120,000	74,400	-
Density (g/cm <sup>3</sup> )	0.97	1	-
105° Residual (%)	11.6	5	98.2
505° Residual (%)	2.5	0.5	3.64
Total Volatile Solid (%)	9.1	4.5	80.9
NH <sub>4</sub> (mg/L)	1,400	78	-
Volatile Fatty Acids (mg/l)	0	0	-
Sulfides (H <sub>2</sub> S) (mg/l)	0	0	-
Alkalinity (meq/l)	140	NA	-
Calorific value (kcal/kg)			3,460

410 Table 1 Physical and chemical parameters of used biomasses.

411

Parameter	Outlet mixture 128 l Digester Co-digestion Test	Outlet mixture 128 l Digester DMY Test
pH	7.24	7.75
BOD 5 (mgO <sub>2</sub> /l)	8,400	1,050
COD (mgO <sub>2</sub> /l)	52,000	33,000
Density (g/cm <sup>3</sup> )	0.97	1
105° Residual (%)	4.8	2.5
505° Residual (%)	1.3	1.2
Total Volatile Solid (%)	3.5	1.4
NH <sub>4</sub> (mg/l)	1,100	1,800
Volatile Fatty Acids (mg/l)	0	0
Sulfides (H <sub>2</sub> S) (mg/l)	0	0
Alkalinity (meq/l)	140	230

412 Table 2 Physical and chemical parameters of co-digestion test.  
413

Parameter	128 l digester Co-digestion test (Average)	128 l digester DMY test (Average)
OLR (g VS/(l*d))	6.35	1.1
HRT (d)	44	32
Specific bio-methane production (l CH <sub>4</sub> /kg VS)	241	219
Biogas quality (%)	54.1	53.6
Biogas yield (l/kg VS)	445	407
Electricity (kWh t/d)	2.9	0.9
Bio-methane Yield (BM <sub>Y1</sub> )	78	

414 Table 3 Comparison of main process parameters.

415

Parameter	128 l digester Co-digestion test (Average)	128 l digester DMY test (Average)
Methane (CH <sub>4</sub> ) (% V/V)	54.1	53.6
Carbon dioxide (CO <sub>2</sub> ) (% V/V)	44.9	45.6
Oxygen (O <sub>2</sub> ) (% V/V)	0.1	0
Carbon monoxide (CO) (ppm)	615.1	287
Hydrogen sulphide (H <sub>2</sub> S) (ppm)	362.5	215

416 Table 4 Biogas average parameters for the conducted experiment.  
417

Parameter	128 l digester Co-digestion test (Average)	128 l digester DMY test (Average)
HRT (d)	44	32
COD feeded (g/l)	110	52
OLR (g)	137.5	89.3
COD reduction (%)	52.7	36.5

418 Table 5 COD behaviour during the experiment.  
419

420 Figures caption:

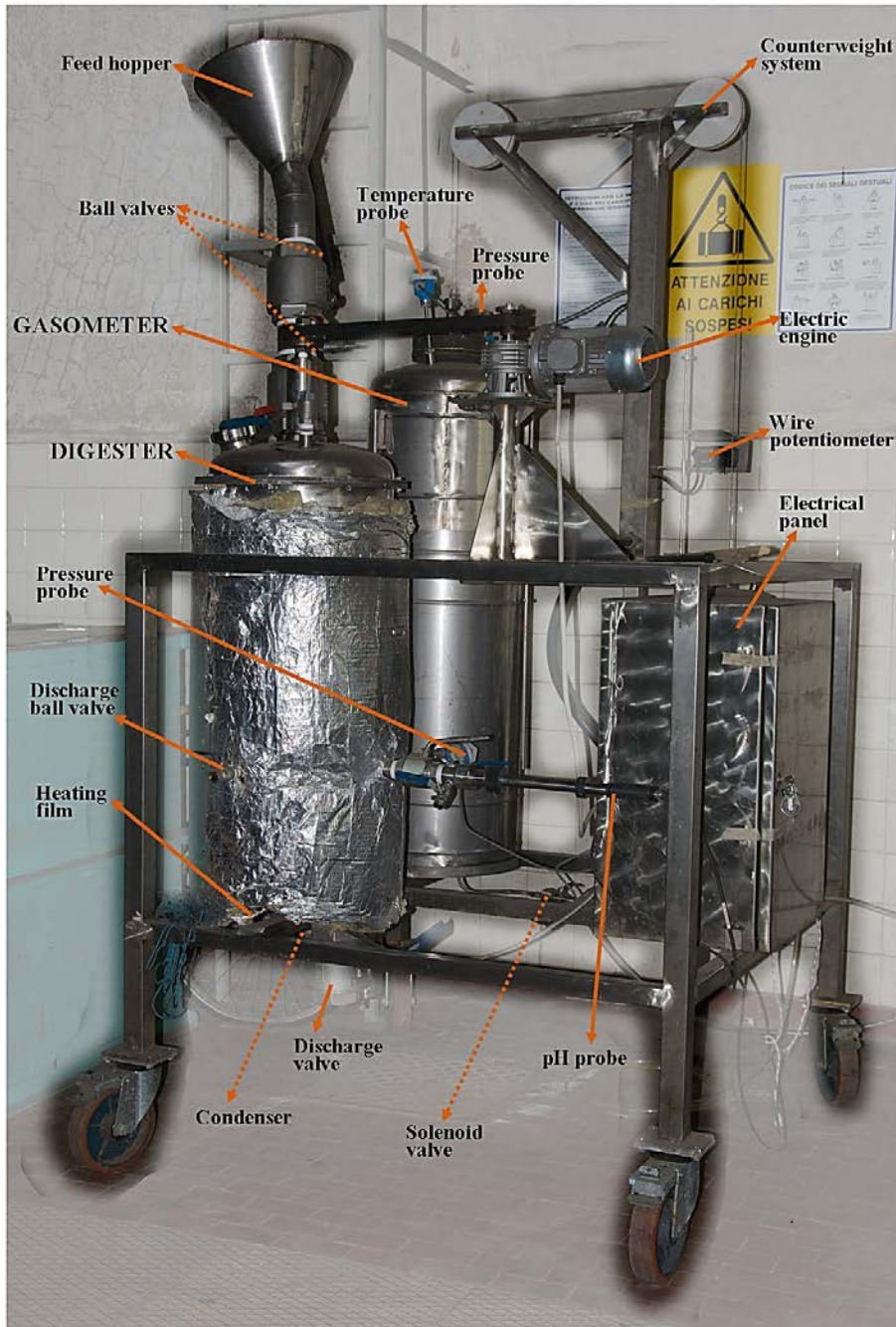
421 Figure 1: Details of the own made experimental 128 L anaerobic digester used in the test.

422 Figure 2: Morgex hybrid phytoremediation plant general layout with details of functional  
423 blocks.

424 Figure 3: Process performance during anaerobic startup, co-digestion and digestate methane  
425 yield test inside the 128 l digester. The cumulative curve indicates the biogas production  
426 during the whole test and the biogas quality curve shows the methane percentage inside the  
427 produced biogas.

428 Figure 4: a) daily biogas production during the startup test; b) during the co-digestion test; c)  
429 during the DMY test.

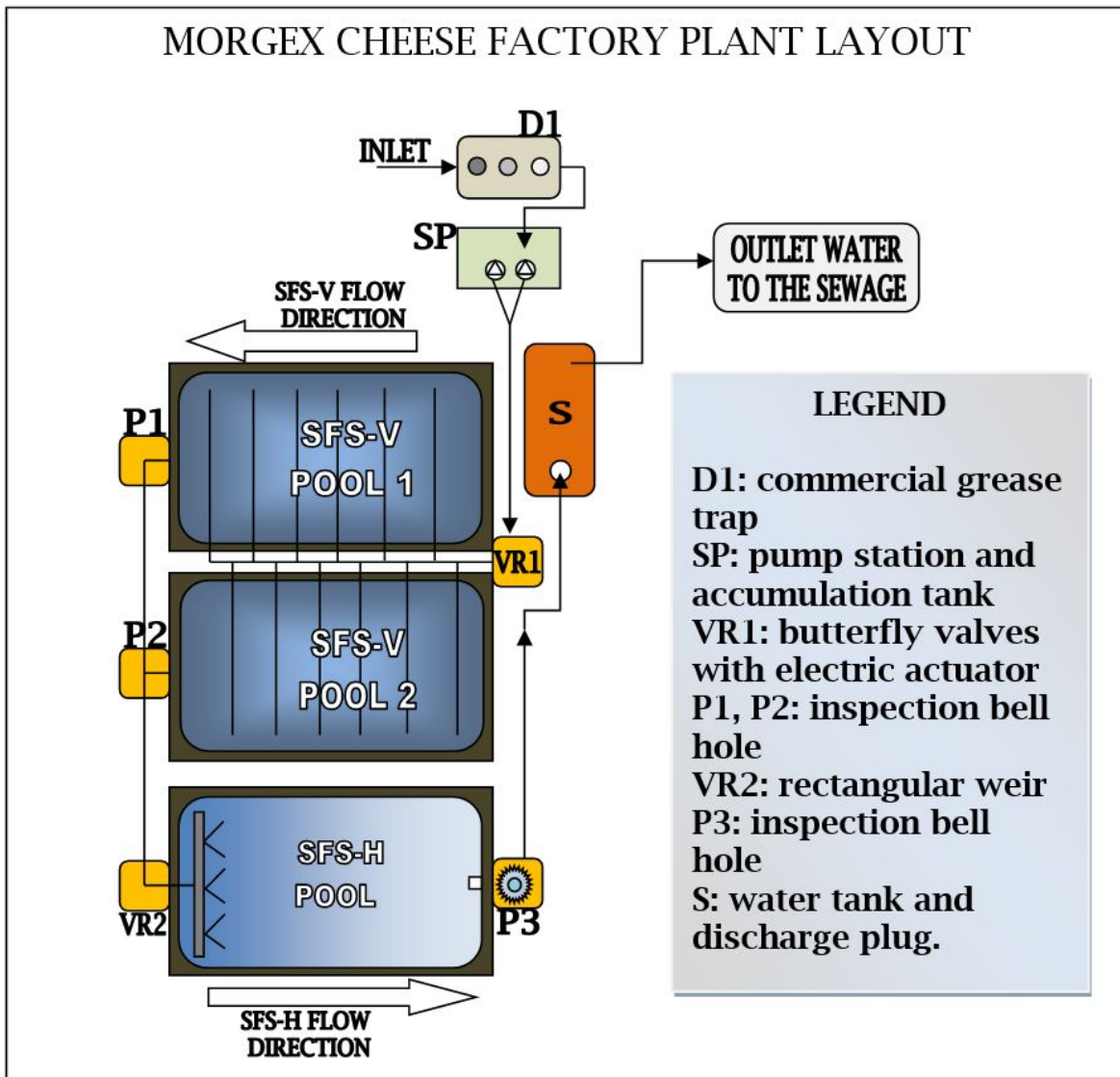
Figure 1





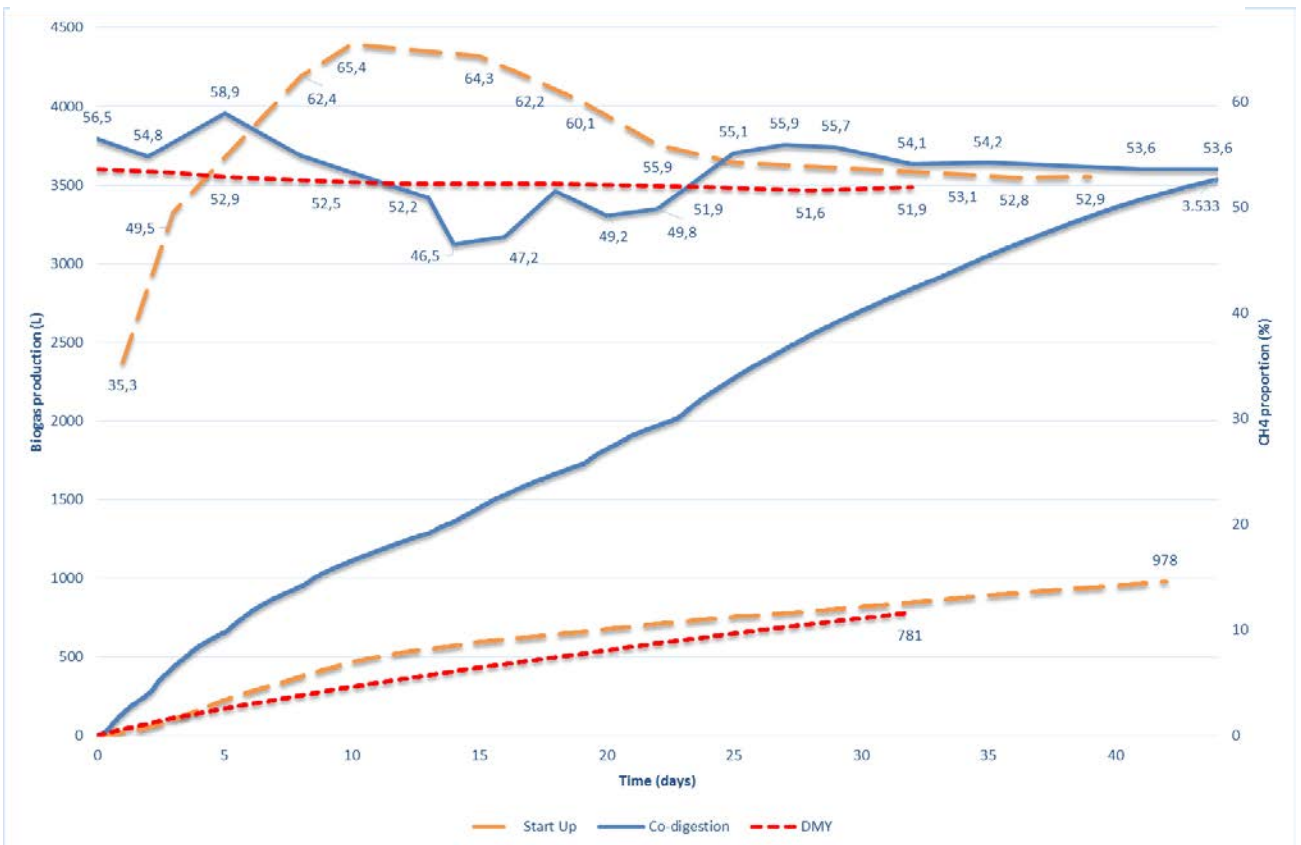
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432 *Figure 2*



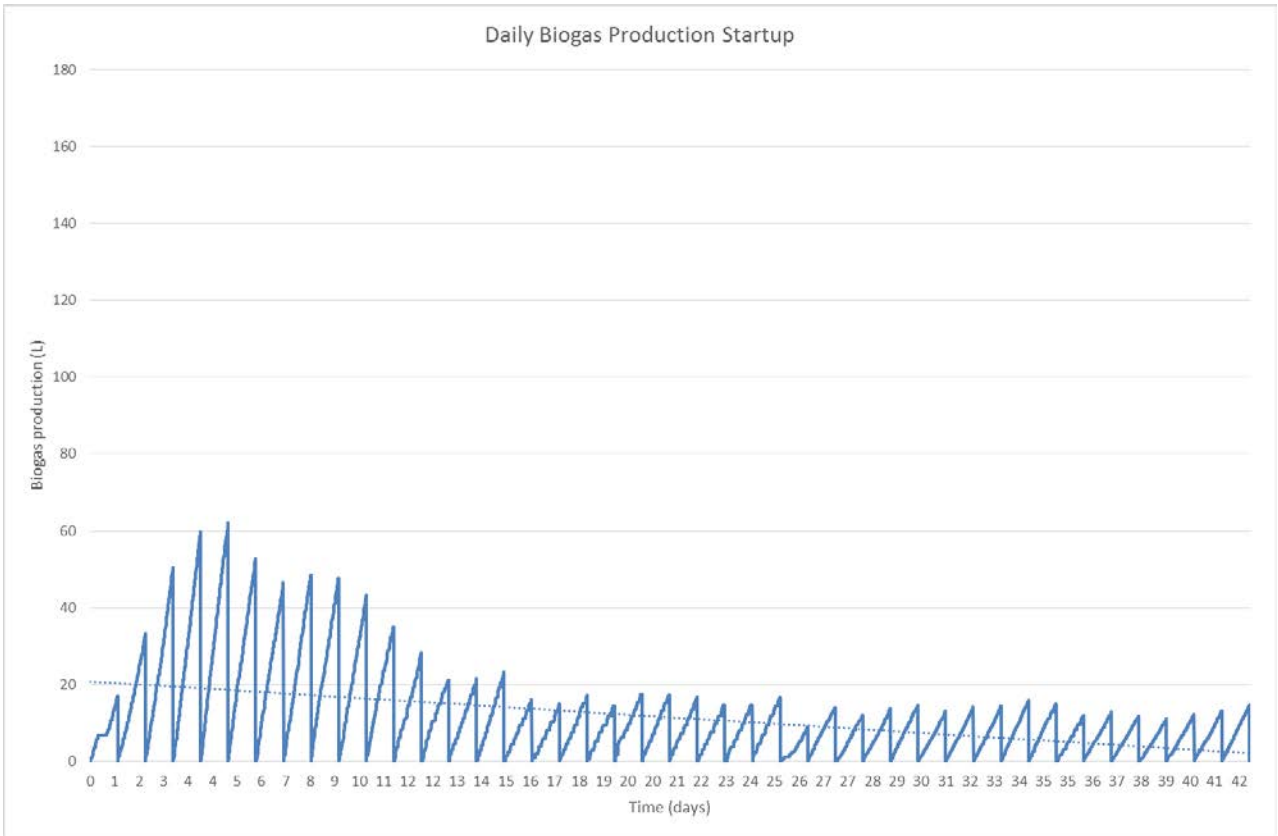
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Figure 3



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Figure 4a



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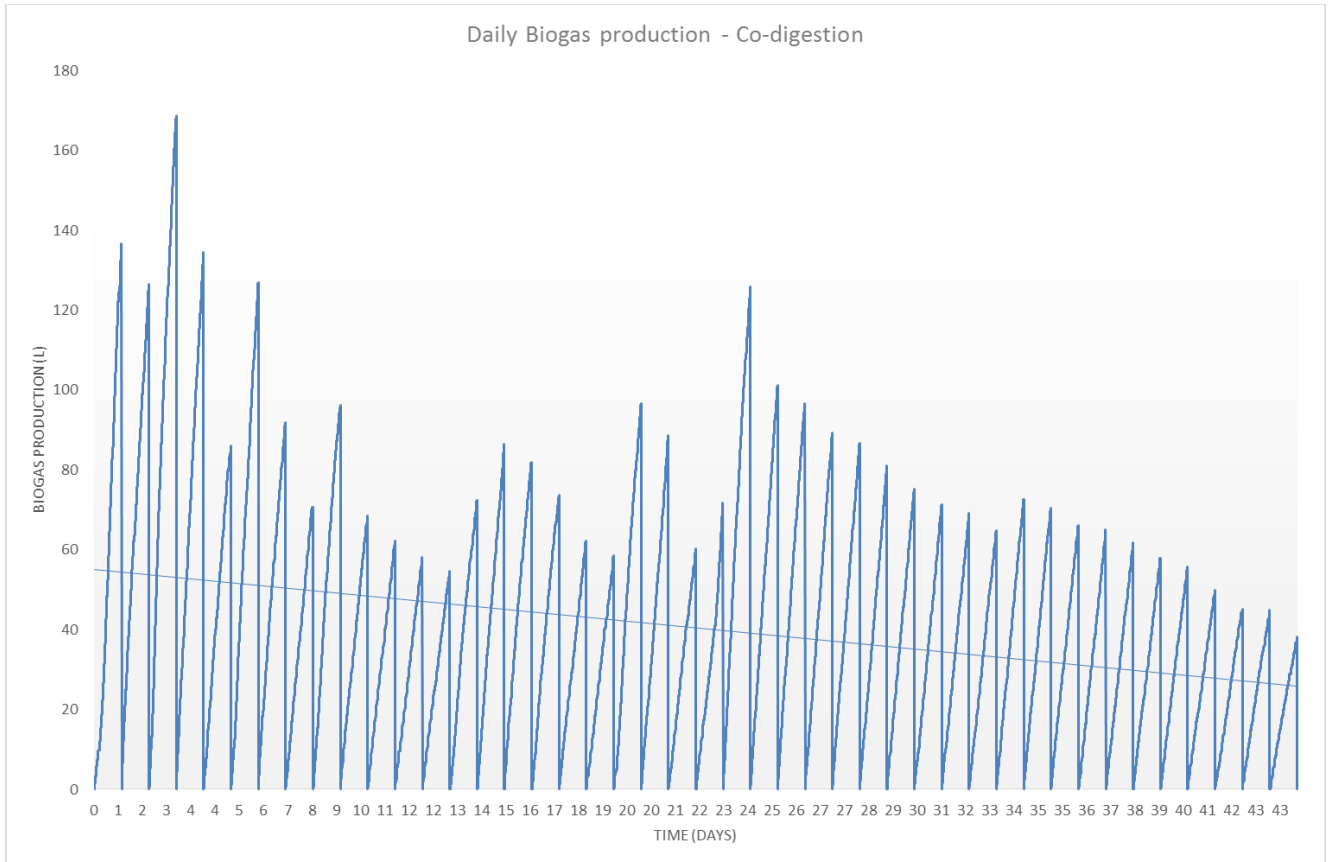
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Figure 4b



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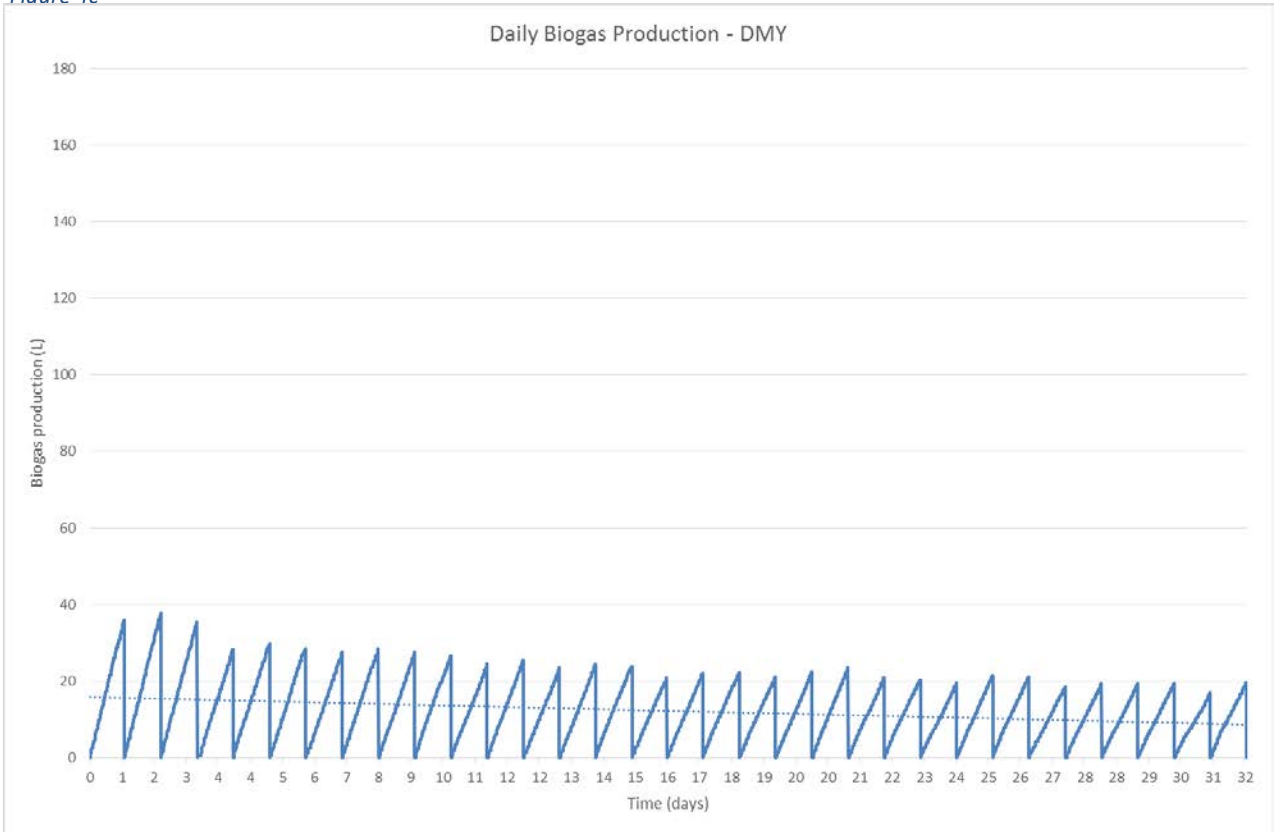
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Figure 4c



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