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High-solids anaerobic mono-digestion of riverbank grass under thermophilic conditions

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ABSTRACT

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The purpose of this study was to investigate the potential of high-solids anaerobic

mono-digestion of riverbank grass under thermophilic conditions, focusing on the effects of

the strength and the amount of inoculum. Ensiled grass was inoculated with three different

inocula; inoculum from liquid anaerobic digester (LI), inoculum from dry anaerobic digester

(DI), and mixture of LI and DI (MI), at feedstock-to-inoculum ratio (FIR) of 1, 2 and 4. The

ensiling process of riverbank grass reduced moisture content (p > 0.05), while the

hemicellulose content was significantly increased from 30.88% to 35.15% (p < 0.05), on dry

matter basis. The highest methane production was at an FIR of 2 with MI (167 $L/kg VS_{added}$),

which was significantly higher (p < 0.05) than with DI, but not significant compared to LI

(p > 0.05). At an FIR of 4, digesters inoculated with LI and DI failed to produce methane,

whereas 135 L_{CH4} /kg VS_{added} was obtained with MI. The kinetic studies showed that at an

FIR of 1 with LI and MI, the inoculum had less of effects on the hydrolysis rate constant

 $(0.269 \text{ day}^{-1} \text{ and } 0.245 \text{ day}^{-1})$ and methane production (135 versus 149 L/kg VS_{added}); rather, it affected the lag phase. In a thermophilic HS-AD of riverbank grass, the mixture of

inoculum with low and high total solids content (TS) helps increase the TS of inoculum and

digestion process. An FIR of 2 was deducted to be the limit for a better startup time and

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48 Introduction

49 Cellulosic biomass is an essentially inexhaustible source of raw material; with an annual production of 1.5 trillion tons 50(Kumar et al., 2008), which can be used for sustainable 51production of environmentally friendly bioenergy such as 52biogas. Among feedstock, it is of interest for bioenergy 53 production because it does not compete with human food or 54animal feed (Yang et al., 2015). Composed mainly of cellulose 55and hemicellulose, and small quantity of lignin (5%-7%) Q2

(Frigon and Guiot, 2010), cellulosic biomass is a good feedstock 57 for anaerobic digestion. Based on total solids content (TS), 58 anaerobic digestion (AD) can be categorized as liquid AD 59 (L-AD) when the TS is lower than 15%; otherwise, it is 60 categorized as high-solids AD (HS-AD) (Li et al., 2011a, b). Q3 Both systems have their own advantages and disadvantages; 62 however, the choice of either L-AD or HS-AD is principally 63 based on the characteristics of the feedstock. For example, for 64 substrates that are low in TS, such as animal manure, sewage 65 sludge and food waste, L-AD is preferable, whereas HS-AD is 66

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higher volumetric productivity of methane.

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more attractive for OFMSW and grass (Li et al., 2011a, b;
Kothari et al., 2014) because they tend to scum, float and
stratify during L-AD (Chanakya et al., 1999).

Riverbank grass is among the cellulosic biomass that 70 is widely available during summer and fall in Japan. For 71 example, on the bank of the Satsunai River, Obihiro, the 72yearly production of grass is estimated to be 600 tons. Generally, 73 it is cut, dried, rolled and incinerated, which obviously causes 74 75 adverse effects on the environment. Therefore, the use of this 76 grass for HS-AD may help recover the energy contained in the grass as a form of methane gas and a valuable compost-like 77 effluent as a biofertilizer. However, it is a challenging process 78 because at a TS of more than 12%, the biogas production of 79 mono-fermentation of ensiled grass, under mesophilic condi-80 tions, drops remarkably, as reported by Koch et al. (2009). 81

Among the disadvantages of HS-AD are its requirements 82 for higher quantities of inoculum and longer hydraulic reten-83 tion times (Li et al., 2011a, b). The strength and amount of 84 inoculum have been reported to have a remarkable impact on 85 the overall performance of the AD process (Forster-Carneiro et 86 al., 2007; Pozdniakova et al., 2012; Dechrugsa et al., 2013). In the 87 case of mono-digestion of grass, which has a low buffer 88 capacity, high C/N ratio and low macro- and micro-nutrient 89 04 contents (Wilkie et al., 1986; Demirel and Scherer, 2011), some 91 challenges are addressed, such as the longer acclimation period 92 of the inoculum to the substrate, slower startup, and longer 93 retention time. However, with a good quality and appropriate 94 quantity of inoculum, mono-digestion of grass can be made more efficient and stable (Yang et al., 2015). For HS-AD, the 95 solids content in the inoculum is an important parameter for 96 97 obtaining high TS content in the digester when it is mixed with the substrate. Up to now, many laboratory works on HS-AD 98 have used centrifuged inoculum from L-AD to inoculate HS-AD 99 of different types of feedstock (Li et al., 2013; Shi et al., 2013; 100 Motte et al., 2013). The main objective of centrifugation is to 101 increase the TS content of the inoculum to obtain a high TS 102content in the digester (Chen et al., 2014), which may affect the 103 microbial community and the macro- and micro-nutrient 104 contents in the inoculum. Another alternative for increasing 105the TS of the inoculum is through the mixture of two different 106 inocula, such as low TS inoculum originating from the L-AD 107 108 process and high TS inoculum originating from the HS-AD process. The effect of this mixture inoculum on methane 109production for HS-AD is investigated and reported on in this 110study. 111

The feedstock-to-inoculum ratio (FIR) has been reported 112 to have a remarkable influence on methane production 113(Dechrugsa et al., 2013; Raposo et al., 2009; Kawai et al., 1142014). It is strictly dependent on substrate characteristics and 115operating temperature (Li et al., 2011a, b; Zhu et al., 2014). The 116 117amount of inoculum, on a volatile solids (VS) basis, is very important in assuring a good startup of AD that helps prevent 118 acidic conditions in the digester (Angelidaki et al., 2009). In 119 120 some cases, inoculum is the only source of nitrogen, heavy 121 metals and the microbial population that guarantees a balanced microbial community in the digester (Zhu et al., 1222014; Xu et al., 2013), especially for high C/N ratio substrates. 123For HS-AD of lignocellulosic biomass, the start-up phase is 124 strictly dependent on the FIR (Motte et al., 2013). In many 125cases, a high FIR is related to an overloading of the feedstock 126

that leads to the accumulation of volatile fatty acids (VFAs) 127 and to acidic conditions in the digester (Dechrugsa et al., 2013; 128 Shi et al., 2014). In contrast, a low FIR is favorable for a faster 129 startup and increased process performance (Motte et al., 2013; 130 Cui et al., 2011). However, lowering the FIR reduces the 131 reactor's efficiency, which can be defined as the methane 132 production per reactor volume (Li et al., 2013). Therefore, 133 investigating the optimum FIR is of interest for increasing the 134 process performance of HS-AD of riverbank grass. 135

In HS-AD, digestion temperature is an important factor as 136 it determines the design of digester. For example, Dranco and 137 Kompogas are operated under thermophilic condition, whereas 138 Valorga is operated under mesophilic condition (Karthikeyan Q5 and Visvanathan, 2013). It is believed that thermophilic condition 140 is more reliable for HS-AD over mesophilic condition because 141 it enhances the AD process, increases digester's efficiency. 142 Although, higher input energy is required under thermophilic 143 condition, the energy balance can be offset by the higher biogas 144 yield under this condition (Li et al., 2011a, b; Karthikeyan and 145 Visvanathan, 2013). Therefore, the main objective of the study Q6 was to investigate the potential of riverbank grass for methane 147 production under thermophilic HS-AD conditions. In particular, 148 the study focused on the effects of the inoculum mixture and its 149 quantity on methane production. Therefore, the specific objec- 150 tives were (1) to determine the best inoculum for HS-AD of 151 riverbank grass and (2) to determine the appropriate riverbank 152 grass and inoculum ratio for methane production. Since river- 153 bank grass is seasonally available substrate, ensiling is an 154 important conservation process to preserve the energy 155 content of the grass and ensure a constant supply for AD 156 plants (Vervaeren et al., 2010; McEniry et al., 2014). Therefore, 157 the effect of ensiling process on the characteristics of grass 158 was also investigated. 159

1. Materials and methods

1.1. Feedstock preparation

Grass was collected in October 2013 on the bank of the 163 Satsunai River, Obihiro, Japan (42°55′N, 143°12′E). Fresh 164 grass was chopped into 20-mm lengths and then 500 g of 165 Si-Master-LP (Snow Brand Seed Co. Ltd., Japan), fermentative 166 lactic acid bacteria composed of Lactococcus lactis and Lactoba- 167 cillus paracasei, per ton of grass was added to ensure the lactic 168 fermentation of the grass. The grass was fermented under 169 anaerobic conditions for 3 months under room temperature. 170 Since inception of experiment was several months after 171 ensiling process, the ensiled grass was kept in a freezer at - 172 20°C until use in order to prevent any change in terms of 173 characteristics and components. Before use, the silage was 174 thawed at 4°C for 24 hr and was air-dried until the moisture 175 content was lower than 10% in order to reduce the specific 176 energy requirements for milling (Barakat et al., 2013). The 177 dried silage was coarse-milled using a centrifugal mill to pass 178 through a 1-mm sieve, which is the optimum particle size to 179 make AD economically viable (Barakat et al., 2013) and 180 enabled more accessible surface for microbial attack (Li et al., 181 2011a, b). The dried silage was kept in airtight bags at room 182 temperature prior to use. 183

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