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1	An Advanced Micro-Bio-Loop to Produce Biogas
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13	Abstract: The authors report an advanced micro-bio-loop that involves recycling through
14	four steps; namely: microalgae culture; de-oxygenation; anaerobic digestion; and aerobic
15	decomposition. The advanced micro-bio-loop operates under sunlight to produce a continuous
16	stream of biogas without requiring any additional external input or internal output to its
17	surrounds. In comparison to conventional biogas production process, it achieves a net positive
18	energy balance at remarkably different level of 0.0224 kWh MJ <sup>-1</sup> , with less than 33% of
19	environmental impacts, less than 0.57% of water demand, only 7.35% arable land-use and
20	0.041% of labor.
21	
22	Highlights

- An advanced micro-bio-loop to produce biogas is proposed.
- The micro-bio-loop can break the bottlenecks of conventional biogas process.
- The overall performance is significantly improved by symbiotic coupling.

26

27 Keywords: biogas, microalga, biomass, biofuel, solar energy utilization

#### 1 **1. Introduction**

Biogas has outstanding versatility and high energy content. As a result, biogas presents an
opportunity to reduce fossil-fuel dependence while contributing significantly to the
sustainable development of rural areas. However, conventional biogas production (Fig. 1(a))
is often limited by problems related to the procurement and pre-treatment of feedstock, the
post-treatment of digestate, and the storage and transportation of both feedstock and digestate
(Lukehurst et al., 2010).

8 To overcome these problems, the authors propose and patent an advanced micro-bio-loop (CN103290059A) that involves recycling through four steps; namely: microalgae culture; 9 10 de-oxygenation; anaerobic digestion; and aerobic decomposition (Fig. 1(b)). The advanced micro-bio-loop operates under sunlight to produce a continuous stream of biogas without 11 12 requiring any additional external input or internal output to its surrounds. Our analysis demonstrates that the micro-bio-loop is much more energy-efficient and more competitive 13 technologically, environmentally, and economically than an equivalent conventional biogas 14 15 production system. These advances imply that the micro-bio-loop has succeeded in breaking 16 the conventional biogas production mode, altering it from "open" to "closed", "complex" to "simple" and "sensitive" to "stable", with the potential to underpin a burgeoning, future 17 biogas industry. 18

### 19 2. Technological description of the advanced micro-bio-loop

20 2.1 Microalgae culture

Single-celled microalgae biomass, promoted as an ideal third-generation biofuel feedstock 21 22 (Cuellar-Bermudez et al., 2015), is employed in the advanced micro-bio-loop. Its productivity offers 10~20 times more biomass yield than terrestrial crops - after all, the mass of 23 microalgae can double in as little as 24 hours (Clarens et al., 2010; Luque, 2010). Microalgae 24 culture production can take place almost everywhere, obviating pressure on arable land, and 25 26 thus greatly reducing food versus fuel concerns (Ashokkumar et al., 2015; Monari et al., 2016). The micro-bio-loop circumvents the need for continuous feedstock procurement, 27 storage, and transportation. Microalgae suspension (which generally contains 0.5 g L<sup>-1</sup> dry 28 29 microalgae biomass) can easily reach a COD value 3 times higher than the industrial design threshold of 500 mgCOD L<sup>-1</sup> (Milieuhygiëne and Foundation, 1989), and so anaerobic 30

1 digestion can be undertaken directly without energy-intensive de-watering and concentration

2 pre-treatment steps.

3 2.2 De-oxygenation

The level of dissolved oxygen (DO) in microalgae suspension can easily reach up to five times 4 the air saturation value (7 mgDO  $L^{-1}$ ) (Jiménez et al., 2003; Mendoza et al., 2013), which is 5 much higher than the methanogenic limit of 0.1 mgDO L<sup>-1</sup> according to Deubling and 6 Steinhauser (2011). For this reason, removal of dissolved oxygen is a critical procedure for the 7 8 next stage of anaerobic digestion. Various liquid-phase de-oxygenation techniques have been 9 employed in industry, including mechanical de-aeration, membrane-based de-oxygenation, heating and chemical reduction (using a deoxidizer), etc. Crucially, these technologies are too 10 costly for the advanced micro-bio-loop and even can impede or halt its activity because of the 11 12 invasion of foreign substances related to de-oxygenation. In the advanced micro-bio-loop, dissolved oxygen is progressively depleted by dark respiration of the microalgae without any 13 addition of deoxidizer, thereby creating an environmentally-stable system. This 14 de-oxygenation method can spontaneously and rapidly generate an oxygen-free microalgae 15 16 suspension, provided the retention time lasts only  $2 \sim 3$  hours. This substantially reduces the capital and operational cost, the latter through a major gain in efficiency. 17

18 2.3 Anaerobic digestion

Conventional feedstock entering an anaerobic digester includes large organic polymers and 19 20 recalcitrant materials that inevitably lower the conversion efficiency and generate inert residues of digestate (Chen et al., 2008). Owing to the absence of lignin, etc. 21 22 (González-Fernández et al., 2012; Popper et al., 2011), microalgae are recognized to be an attractive substrate for anaerobic digestion, noting that microalgae produce  $0.53 \sim 0.80$  LCH<sub>4</sub> 23 g<sup>-1</sup>VS (González-Fernández et al., 2012) (i.e. liters of methane per gram volatile solids). The 24 resultant digestate does not contain inert residues, and so can achieve full recycling whereby 25 26 fertilizer is created for the microalgae culture, accordingly reducing the disposal problem. The proportion of methane in the biogas produced lies in a similar range (i.e.  $60\% \sim 75\%$ ) to that 27 28 of the majority of other microalgae-based studies related to biogas applications, regardless of 29 species and operating conditions (Ras et al., 2011).

30 2.4 Aerobic decomposition

1 Digestate is a highly valuable fertilizer which, if used effectively, can significantly offset inorganic fertilizer. However, conventional biogas production usually involves pumping 2 digestate and then spreading it on nearby land or discharging into receiving waters without 3 proper post-treatment. This practice greatly increases the risks of lake eutrophication arising 4 from N and P outputs, the spread of pathogens from one farm to another, and the release of 5 contaminants into the food chain. In the advanced micro-bio-loop, reuse of a digestate 6 suspension to cultivate microalgae not only lowers the need for chemical fertilizer to promote 7 8 microalgae production but also avoids the aforementioned risks. Moreover, water in the 9 digestate suspension can also be re-used simultaneously in further microalgae cultivation. However, the digestate suspension contains some organically-bounded nutrients that are 10 generally believed to be indigestible by or even toxic to the dominant microalgae species in 11 12 nature (Uggetti et al., 2014). For direct feed back into the microalgae culture without pre-treatment, the resulting recycling ratio of partial nutrients will merely remain unaltered at 13 about 50~80% (Brennan and Owende, 2010; Golueke and Oswald, 1959). In order to 14 achieve a more optimal recycling ratio and keep microalgae at their best, advantage is taken 15 16 of aerobic bacteria mineralization to convert organic nutrients in the digestate into inorganic forms that are readily digestible by microalgae. The nutrients and water in the advanced 17 micro-bio-loop correspond to recycling ratios that are almost always 100%, achieving in situ 18 quality management of the digestate. Consequently, the fertilizer cost substantially decreases 19 from 0.012 \$ kg<sup>-1</sup>algae year<sup>-1</sup> to 0 \$ kg<sup>-1</sup>algae year<sup>-1</sup> (under 25 g algae m<sup>-2</sup> d<sup>-1</sup> productivity), 20 21 which accounts for 30% cost and 45% effective energy of the microalgae culture (Clarens et 22 al., 2010).

In the present study, the advanced micro-bio-loop employed naturally dominant species, i.e., aerobic and anaerobic bacteria taking from domestic sewage treatment plant, and microalgae taking from natural local lake. Tab.1 shows the mass and energy flows of the advanced micro-bio-loop with the considered functional unit of 890 MJ, produced by the combustion of CH<sub>4</sub> in an internal combustion engine.

# **3.** Comparison of the advanced micro-bio-loop with conventional biogas production

29 from the perspective of sustainability

30 Life Cycle Sustainability Assessment (LCSA) is performed to analyze potential impacts of the

1 advanced micro-bio-loop and conventional biogas production process typically on natural 2 resources, human society, economy and environment. The life time of processing infrastructures is assumed to be 15 years. The advanced micro-bio-loop considered herein is 3 located in the middle of China. The relevant analysis on it is based on our experimental 4 observation and material balance. The inventory of conventional biogas production process is 5 derived from academic resources, engineering design standards, communications with 6 7 industrial producers, and processes described in the Ecoinvent Database. (See Supplementary 8 Table S1-S6)

Advanced micro-bio-loop and conventional biogas production process achieve a net positive 9 energy balance at remarkably different levels, 0.0224 kWh MJ<sup>-1</sup> and 0.0539 kWh MJ<sup>-1</sup>. The C, 10 N, P, K, and water balances in the micro-bio-loop corresponds to recycling ratios of 103.4  $\pm$ 11 12 0.5%, 99.8±1.9%, 102.7±1.1%, 104.2±0.9% and 99.97% respectively, achieving a free supply of fertilizer and water. Fig. 2 compares the impacts generated by producing 1 MJ biogas using 13 these two systems from the perspective of Life Cycle Sustainability Assessment. Each impact 14 is standardized according to the value of the worst scenario specific to the impact. The results 15 16 indicate that the micro-bio-loop is vastly preferable to conventional biogas production in terms of energy use, acidification potential, global warming potential, ozone layer depletion, 17 eutrophication, total investment cost, water demand, arable land use, and labor. All the 18 preceding impacts are less than 33% of conventional biogas production. The water demand of 19 the micro-bio-loop  $(3.66 \times 10^{-5} \text{ m}^3 \text{ MJ}^{-1})$  is less than 0.57% of conventional biogas production 20  $(6.42 \times 10^{-5} \text{ m}^3 \text{ MJ}^{-1})$  at the same functional unit. Due to the particular feedstock and simple 21 infrastructure inherent to the micro-bio-loop system, its requirements for arable land-use and 22 labor are only 7.35% and 0.041% of conventional biogas production. From the above insights 23 24 into technological, environmental and economic dimensions, the advanced micro-bio-loop has much competitive performances beyond an equivalent conventional biogas production 25 26 system.

In China, particularly in rural areas, the enthusiasm for household biogas production schemes has waned from an initial flourish of interest, partly because of a lack of scientific management by farmers of the inputs and outputs, along with a lack of inspection and maintenance of equipment. The advanced micro-bio-loop has more than

\$40 billion market potential to revitalise household biogas production and could
 simultaneously mitigate at least 210 million tons CO<sub>2</sub> emission over the next 5 years,
 thereby making great contributions to China's Renewable Energy Development Plan
 and China's National Plan for Climate Change (2016-2020) (Committee, 2007, 2014).

#### **4.** A more efficient improvement for the advanced micro-bio-loop industrialization

6 Vigorous aeration is required to provide sufficient CO<sub>2</sub> for the microalgae culture and O<sub>2</sub> for aerobic decomposition (Grady Jr et al., 2011; Richmond, 2008). The associated energy 7 8 consumption proves to be a millstone, resulting in an operational cost increase of 20%. In fact, 9 only a coupled reactor can substitute for aerators in the advanced micro-bio-loop, without requiring any energy input, noting that both microalgae and aerobic bacteria exhibit a high 10 degree of uniformity in the survival environment due to their symbiotic behavior. More 11 12 precisely, microalgae produce the  $O_2$  necessary for aerobic bacteria to mineralize organic matter, consuming in turn the CO<sub>2</sub> released by respiration of the aerobic bacteria (Praveen and 13 Loh, 2015). According to the classical two-film theory, the CO<sub>2</sub>/O<sub>2</sub> transfers involved in the 14 microalgae culture and aerobic decomposition processes are all essentially controlled by 15 16 liquid-films and gas-films (Fig. 3(a) and Fig. 3(b)). Use of a coupled reactor can completely eradicate the resistances of the four films created by microalgae culture and aerobic 17 decomposition (Fig. 3(c)), thereby encouraging smoother exchange of  $CO_2$  and  $O_2$  and 18 providing a mechanism for efficient mixing. 19

#### 20 **5.** Conclusions

micro-bio-loop involves a completely 21 The advanced independent, stable micro-ecosystem, which comprises a sustainable cycling eco-chain of producers, 22 consumers, and decomposers (microalgae, anaerobic bacteria, and aerobic bacteria). 23 24 The resulting micro-ecosystem differs to that of a conventional biogas production process in that the former simultaneously achieves full internal and external 25 26 circulations of all substances. Technological simplicity combined with full circulation at steady-state operation make the advanced micro-bio-loop a particularly attractive 27 28 option due to its large scale availability, tractable technology, ease of installation, safe 29 operation, and overall economic efficiency. The advanced micro-bio-loop could

- 1 progressively substitute for a significant proportion of biogas energy production
- 2 facilities, with significant societal benefits.
- 3

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- 6

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#### **Figure captions**

Fig. 1 Schematic diagrams of: (a) conventional biogas production process; and (b) advanced micro-bio-loop.

Fig. 2 Comparison of impacts generated by production of 1 MJ biogas using the conventional biogas production system and the advanced micro-bio-loop. The labels GWP, AP, EP, ODP, Arable land, Water, Labour, and Expense refer to global warming potential, acidification potential, eutrophication potential, ozone depletion, arable land use, water consumption, labour cost, and total investment.

Fig. 3 Two-film theory models of: (a) microalgae culture; (b) aerobic bacteria culture; and (c) microalgae coupled to aerobic bacteria culture.

# Table 1 Mass and energy flows of the advanced micro-bio-loop with the functional unit

of	890	MJ
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Туре	Description	Quantity	Units			
Operatio	n 1: microalgae culture					
Output	Flow out of the photo-bioreactor	310.288	m³			
Input	Mineralized digestate (contains inorganic N, P and K)	310.254	m³			
Input	Ν	0.000	kg			
Input	Ρ	0.000	kg			
Input	К	0.000	kg			
Input	Water	0.0325	m <sup>3</sup>			
Input	Electricity consumption (air pump)	1.310	kWh			
Input	Electricity consumption (pumping)	0.873	kWh			
Operation 2: de-oxygenation						
Output	Flow out of the de-oxygenation plant	310.288	m <sup>3</sup>			
Input	Flow out of the photo-bioreactor	310.288	m <sup>3</sup>			
Input	Electricity consumption (pumping)	3.939	kWh			
Operatio	n 3: anaerobic digestion					
Output	Biogas (70% CH <sub>4</sub> )	32.250	m <sup>3</sup>			
Output	Digestate (contains organic N, P and K)	310.254	m <sup>3</sup>			
Input	Electricity consumption (pumping)	3.939	kWh			
Input	Heat consumption (internal biogas)	2.047	kWh			
Input	Flow out of the de-oxygenation plant	310.288	m <sup>3</sup>			
Operation 4: aerobic decomposition						
Output	Mineralized digestate (contains inorganic N, P and K)	310.254	m <sup>3</sup>			
Input	Digestate	310.254	m <sup>3</sup>			
Input	Electricity consumption (air pump)	1.310	kWh			
Input	Electricity consumption (pumping)	0.873	kWh			
Operation 5: purification						
Output	Methane, 96%	22.575	m <sup>3</sup>			
Input	Biogas (70%CH <sub>4</sub> )	32.250	m <sup>3</sup>			
Input	Electricity consumption	4.291	kWh			
Operation 6: combustion						
Output	Energy (from methane)	172.000	kWh			
Input	Methane, 96%	22.575	m <sup>3</sup>			



Fig. 1 Schematic diagrams of: (a) conventional biogas production process; and (b)

advanced micro-bio-loop.



Fig. 2 Comparison of impacts generated by production of 1 MJ biogas using the conventional biogas production system and the advanced micro-bio-loop. The labels GWP, AP, EP, ODP, Arable land, Water, Labour, and Expense refer to global warming potential, acidification potential, eutrophication potential, ozone depletion, arable land

use, water consumption, labour cost, and total investment.



Fig. 3 Two-film theory models of: (a) microalgae culture; (b) aerobic bacteria culture;

and (c) microalgae coupled to aerobic bacteria culture.