

## A Review on Application of Microorganisms for Organic Waste Management

Chee Woh Leow<sup>a</sup>, Yee Van Fan<sup>b</sup>, Lee Suan Chua<sup>c</sup>, Ida Idayu Muhamad<sup>a</sup>, Jiří Jaromír Klemeš<sup>b</sup>, Chew Tin Lee<sup>a,\*</sup>

<sup>a</sup>Faculty of Chemical and Energy Engineering, Universiti Teknologi Malaysia, 81310 Johor Bahru, Johor, Malaysia

<sup>b</sup>Sustainable Process Integration Laboratory – SPIL, NETME Centre, Faculty of Mechanical Engineering, Brno University of Technology - VUT Brno, Technická 2896/2, 616 69 Brno, Czech Republic

<sup>c</sup>Institute of Bioproduct Development Universiti Teknologi Malaysia, 81310 Johor Bahru, Johor, Malaysia  
 ctlee@utm.my

The extensive utilisation of microorganisms namely fungi and bacteria for treating organic wastes has been attributed to their efficiency in eliminating pathogen and accelerating the degradation process. Their uses have been found considerably efficient for enhancing waste treatment. Among many methods employed, composting mediated by indigenous microbial communities has gained significant popularity in treating organic waste. Use of cellulolytic microorganisms to expedite the degradation rate of wastes, notably the lignocellulosic components, may prove useful. This paper reviews the application of microorganisms in the waste management technologies that include anaerobic digestion and composting of organic waste with a high lignocellulosic portion, composting of heavy metal contaminated organic waste, and composting at low temperature.

### 1. Introduction

The rapid population growth, and the increase of municipal solid waste (MSW), agricultural waste and food waste contributed to the emission of Greenhouse Gases (GHG) and challenged the current waste management practices. Composting or anaerobic digestion (AD) of organic waste are alternative solutions instead of direct disposal to landfill which causes an impact on the environment due to the emission of GHG and unpleasant odour (Al Zuahiri et al., 2015).

Organic wastes that are abundant in the organic fraction can be converted into renewable biogas and compost by microorganisms under controlled conditions (Wang et al., 2011). The digestate from the AD system can serve as fertiliser for soil enhancement (Kiran et al., 2014). During agricultural waste composting, lignocellulose, starch, and protein account for the significant part of biomass. The abundant lignocellulose composition often requires pretreatment process such as chemical and industrial enzyme added, thermal treatment or biological treatment using microorganisms before composting or AD. Among the microorganisms, microbes and fungi (MF) are more favourable due to its efficiency in degrading the organic matter (Fan et al., 2017).

Lignocellulose material is the most abundant biomass, yet the degradation rate and biogas production are much lower than other organic waste. It is of great interest to understand the trend on how different types of microbes could effectively treat lignocellulose waste, increase the biogas yield, remediate contaminated organic waste by composting, and facilitate composting at low temperature. This paper reviews mainly the use of bacteria and fungi applicable to the waste treatment system, notably for the composting of agricultural waste and the increment of potential biogas in the AD.

### 2. Composting

The composting process involves three phases, namely mesophilic, thermophilic, and maturation, which uses diverse microflora, such as mesophilic and thermophilic bacteria, fungi and actinomycetes to convert and

stabilise the organic waste to humus (Zeng et al., 2001). The physiochemical condition during various phases, such as oxygen, temperature, moisture content and nutrient availability, determine the development of microbial populations during composting. The microbial secrete different enzymes to hydrolyse the complex organics matter to a stable and simple form and eventually produce a product such as humus and biogas.

Temperature is a significant parameter in the composting process. Composting has a typical temperature profile of a quick increase in temperature of up to 65 or even 80 °C in the first few days. Composting involves a rapid transition from a mesophilic to a thermophilic microbial community and followed by a slow decrease in temperature. As the diversity of microorganisms increases, fungi and mesophilic bacteria re-establish themselves. At the thermophilic stage, thermophilic bacteria can degrade complex material such as lignin, protein, chitin and cellulose.

During the disintegration process, the particulate organic matter was disintegrated into carbohydrates, lipids and proteins and followed by enzymatic hydrolysis to short chained carbohydrates, long chain fatty acids and amino acids (Lauwers et al., 2013). These hydrolytic enzymes including protease, lipases, cellulase and amylase, are secreted by the microorganisms presented in the bulk liquid or attached to particulates (Vargas-Garcia et al., 2010). Challenges remained in the composting process due to high variation of waste composition (Abdullah et al., 2013), long retention/residence time, temperature sensitive (UNEP, 2015), and hygiene concern/odour control (Wang et al., 2003).

For agricultural waste, challenges arise due to the abundant of lignocellulose composition. Acid and thermal pretreatments of lignocellulose are required before composting process. Pretreatment methods using thermal or chemical is not favourable due to energy consumption and impact of added chemicals to environmental (Rouches et al., 2016). Compared to chemical and thermal methods, the use of an enzyme such as cellulase to treat the lignocellulose waste is desirable, the industrial enzyme is costly if applied at large scale. The application of thermophilic cellulolytic microorganisms including fungi and bacteria to expedite the composting process is preferable (Bohacz, 2017). Fungus such as *T. reesei* has been reported to produce more than 100 g of cellulase per L of culture broth and their ability to grow in liquid and solid medium make it a suitable candidate for treating agricultural waste (Schuster and Schmoll, 2010). This finding suggested that application of thermophilic cellulolytic microorganisms in agricultural waste could reduce the dependency on the industrial enzyme, chemical and thermal pretreatments.

There is no universal method for composting as the types of substrate and its physio-chemical condition can influence the process. Co-composting is an integrated sustainable process that offers some advantages over composting that uses a single substrate. Recent studies have focused on co-composting using different types of agricultural waste. Co-composting of food waste with Chinese medicine herbal residues was reported to enhance the anti-pathogenic property in compost. The process inhibits the activities of *Alternaria solani* and *Fusarium oxysporum* that cause early blight and vascular wilt in potato/tomato plants, followed by multiple phytopathogenic infections leading to plant damping-off (Zhou et al., 2016). These findings concluded the potential of sustainable ways to control the soil-borne pathogen instead of using the commercial fungicides that can cause negative effects on the environmental and human health.

Awasthi et al. (2015) studied the effect of various bulking waste such as wood shaving, agricultural and yard trimming waste combined with an organic fraction of municipal solid waste (OFMSW) composting by assessing their influence on microbial enzymatic activities and quality of finished compost. The results suggested that OFMSW combined with wood shaving and microbial consortium (*Phanerochaete chrysosporium*, *Trichoderma viride* and *Pseudomonas aeruginosa*) were the helpful tool to facilitate the enzymatic activity and shortened composting period within four weeks.

Microbial also influence on nitrogen conservation, pH buffering during co-composting and end product quality. Kumar et al. (2013) used gelatine industry sludge (GIS) combined with an organic fraction of municipal solid waste (OFMSW) and poultry waste (PW) that employed zeolite and mixed with the enriched nitrifying bacterial consortium (ENBC). The ENBC is prepared using the functional strains of *Pseudomonas aeruginosa*  $6.5 \times 10^8$  colony-forming units (CFU)/mL, *B. licheniformis*  $7.2 \times 10^6$  CFU/mL, *B. subtilis*  $6.9 \times 10^6$  CFU/mL and *Bacillus cereus*  $7.8 \times 10^5$  CFU/mL, which had been isolated by enrichment method from gelatine wastewater. Their findings suggested that the best mixture contained GIS, OFMSW and PW in a ratio 6 : 1 : 0.5 (dry weight basis), inoculated with 10 % zeolite and ENBC effectively accelerated the composting process and reduced the nitrogen loss, as indicated by the nitrogen dynamics and maturity parameters of the end products (Awasthi et al., 2016).

The effect of *Phanerochaete chrysosporium* (white-rot fungi) inoculation during drum composting of agricultural waste, performed at different composting stages using substrate combination of vegetable waste, cattle manure, sawdust and dried leaves with a total mass of 100 kg in a 550 L rotary drum composter, showed 1.45 fold and 1.7 fold reduction of volatile solids in trial 2 (inoculated at mesophilic phase) and trial 3 (inoculated at thermophilic phase) as compared to trial 1 without the addition of fungal as inoculum (Varma et al., 2015). This finding suggested that inoculation of fungus after the thermophilic phase was found more

effective than inoculation during the initial days of composting for producing more stabilised and nutrient-rich compost. Table 1 shows the application of microorganisms in composting process.

Table 1: Application of microorganisms in composting process

Name	Type/ Source of Microbe	Temperature	Amount & Concentration	Main Substrate	Research Findings	References
Pseudomonas fragi, Mix strain Pseudomonas simiae, Clostridium vincentii, Pseudomonas jessenii and Iodobacter fluviatilis		10 °C	1 x 10 <sup>8</sup> CFU/ mL, 1 % in dry weight	Food waste & maize straw	Contributed to composting start-up at low temperature	Xie et al. (2017)
Brevundimonas diminuta CB1, Flavobacterium glaciei CB23 Aspergillus niger CF5 and Penicillium commune CF8 Pichia kudriavzevii RB1	Psychrotrophic bacteria (isolated from frozen soil) and thermophilic fungi (isolated from compost)	-2 - 5 °C	1 x 10 <sup>8</sup> CFU/ mL, 10 mL/kg	Dairy manure & rice straw	Promotes maturity of dairy manure-rice straw composting under cold climate conditions	Gou et al. (2017)
Phanerochaete chrysosporium	White-rot fungi	25 - 29 °C	2 x 10 <sup>6</sup> CFU/ mL, 2 % in wet weight	Model food waste (commercial rabbit food and cooked rice) Rice straw, bran, vegetables and soil	Promote degradation of organic acid and accelerating the composting process Stabilise in composting of lead-contaminated agricultural waste	Nakasaka and Hirai (2017) Huang et al. (2017)

### 3. Anaerobic Digestion (AD)

Management of organic waste by biogas production provides twofold advantages including GHG minimisation and renewable energy generation. AD is a technology where organic matter is degraded by a consortium of microorganism and transformed into methane-rich biogas as an alternative to natural gas. The resulting effluent can be used for fertiliser production. The energy conversion efficiencies of the AD, particularly for agricultural waste (crop residues), can be limited due to the lignocellulosic composition which is recalcitrant to biodegradation. The potential biogas yield from lignocellulosic biomass (> 100 m<sup>3</sup>/t) (IEA Bioenergy, 2015) is higher than the other type of feedstock such as cattle slurry (15 - 25 m<sup>3</sup>/t) and poultry (30 - 100 m<sup>3</sup>/t) (NNFCC The Bioeconomy Consultants, 2016). Out of the four stages in AD process (hydrolysis, acidogenesis, acetogenesis and methanogenesis), hydrolysis of lignocellulosic biomass has been commonly determined as the primary rate-limiting step (Christy et al., 2014). The theoretical yield based on the cellulose content of agricultural waste was predicted to be about 90 %, but the methane production efficiency is just 50 % due to the inefficient hydrolysis of biomass within full-scale biogas reactors (Azman et al., 2015). The highlighted requirement of long retention time, resulting in the higher capital cost for a larger reactor, minimal energy generation efficiency and less feasible for implementation. The effectiveness of biogas production is usually low without additional substrate treatment before or during AD process. Improving the hydrolysis efficiency of lignocellulosic biomass is in need for drastic improvement of AD implementation. Utilisation of microorganism in the AD could substantially increase the enzyme loading and eventually promote the degradation of lignocelluloses in a cost-effective way.

The common fungi class used for biofuel production is white rot fungi (Poszytek et al., 2016) that prefers the aerobic condition and sterilised condition. Fungal treatment mainly attacks lignin, but microbial consortium

forms of bacteria usually have high and hemicelluloses degradation ability. Peng et al. (2014) stated that the economic feasibility of fungal pretreatment is low due to the loss of polysaccharide components during fungal growth and long cultivation period reduce overall productivity. Table 2 shows the various applications of microorganisms in crop residues (lignocellulosic biomass) for biogas yield enhancement.

*Table 2: Application of microorganisms in crop residues for biogas yield enhancement*

Name	Type/ Source of Microbe	Temperature	Amount & Frequency	Main Substrate of AD	Increment of Biogas Potential	References
Pseudobutyrvibrio xylanivorans Mz 5 <sup>T</sup>	Rumen bacteria, anaerobic	37; 35 °C	5 vol%; Once	Brewery spent grain	17.8 %	Čater et al. (2015)
Pseudobutyrvibrio xylanivorans Mz 5 <sup>T</sup> + Fibrobacter succinogenes S85	Rumen bacteria, anaerobic	37; 35 °C	5 vol%; Once	Brewery spent grain	6.9 %	Čater et al. (2015)
Clostridium cellulovorans 743B	Anaerobic, mesophilic	37; 35 °C	5 vol%; Once	Brewery spent grain	3.9 %	Čater et al. (2015)
Consortium	Predominantly of the genus Clostridium	37 - 40 °C	10 wt%; Daily (routine)	Sweet corn residues	15 % (compare to processing one time), 56 % (compare to non-bioaugmentation)	Martin-Ryals et al. (2015)
Consortium	From compost (Clostridium sp., Pseudoxanthomonas sp., Brevibacillus sp., Bordetella sp.)	50; 35 °C	2 wt%; Once	Maize Straw	74.7 %	Hua et al. (2016)
Consortium	Thermophilic (from soil samples filled with rotten lignocellulosic materials)	55; 55 °C	2 vol%; Once	Cassava residues	96.63 %	Zhang et al. (2011)
Consortium	Yeast, cellulolytic bacteria, lactic acid bacteria	Not reported	0.01 wt%; Once	Corn straw	33.07 %	Zhang et al. (2011)
Isolated from compost (consortium, predominant by Clostridia class)	Anaerobic, thermophilic, cellulolytic,	55; 55 °C	0.1 vol%; Once	Cellulosic substrate (lab test-filter paper)	14.5 %	Kinet et al. (2015)
Isolated from sewage sludge cattle slurry and manure (consortium)	Cellulose degrading bacteria	30; 30 °C	10 % (v/v), pretreatment, different reactor	Maize silage	38 %	Poszytek et al. (2016)

Based on Table 2, a single bacteria strain and consortium are applied in the recent studies to increase the biogas yield. A consortium contributes to a higher yield of biogas production than the single strain. The use of the consortium allows for maintaining a high level of hydrolysis even at the mesophilic temperature. This demonstrated a great advantage in term of energy consumption. It also avoids the problems of feedback regulation and metabolite repression associated with the use of the single isolated strains. The microbial consortium is better adapted to pH and temperature changes and tends to show higher resistance to the presence of heavy metals, toxic organic compounds or contamination by other strains. The sterilisation of lignocellulosic feedstock is not necessary, which could lower costs and save time. The uses of single or mixed strains cultivation in the hydrolysis of lignocellulosic materials are not in accord with the degradation characteristics of lignocelluloses in nature (Zhang et al., 2011). Lignocellulosic materials are degraded under the cooperation of many microorganisms by producing a variety of cellulolytic and hemicellulolytic enzymes. This suggested that consortium is a better option than the isolated single or mixed strains.

Martin-Ryals et al. (2015) suggest a routine bioaugmentation with the cellulolytic microorganism. Bioaugmentation is usually referred as a part of AD process (in the digester) where single or mixed strains are

added (Martin-Ryals et al., 2015) in contrast to the other biological pretreatment (an additional process before the substrate is fed into the reactors). Addition of mixed strains increased the biogas production by 15 % during the AD of sweet corn processing residues as compared to that added with a single strain. The positive effect of the economic analysis and more in-depth calculation are needed to verify the practicality of routine bioaugmentation as the substrate treatment for the AD.

#### 4. Conclusions

Microorganisms including bacteria and fungi have proven to enhance the degradation process based on the previous studies. The application of microorganism consortium in composting and AD provides an alternative solution to waste management as chemical and thermal methods are not favourable in term of cost and energy consumption. Co-composting aided by microorganisms offers the co-benefits of enhanced degradation and minimised valorisation of nutrients in the compost. An increment of biogas potential ranging from 3.9 % to 96.63 % is achievable with the application of microorganisms for crop residues in the AD system. Although the inclusion of microbial could enhance the performance of composting and AD, the economic feasibility of microbial culture cost remains the major concern in future studies.

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