Journal of Engineering Science and Technology
Special Issue on SOMCHE 2014 & RSCE 2014 Conference, January (2015) 10 - 18

EFFECTS OF MICROBIAL ADDITIVE ON THE PHYSIOCHEMICAL AND BIOLOGICAL PROPERTIES OF OIL PALM EMPTY FRUIT BUNCHES COMPOST

LI YEE LIM[1], LEE SUAN CHUA[1,*], CHEW TIN LEE[2]

[1]Metabolites Profiling Laboratory, Institute of Bioproduct Development, Universiti Teknologi Malaysia, 81310 UTM Skudai, Johor, Malaysia
[2]Department of Bioprocess Engineering, Faculty of Chemical Engineering, Universiti Teknologi Malaysia, 81310 UTM Skudai, Johor, Malaysia
*Corresponding Author: chualeesuan@utm.my

Abstract

This study was focused on the effects of microbial additive on oil palm empty fruit bunches (EFB) compost. The physical, chemical and biological properties of the compost during composting were investigated. Significantly, microbial inoculated compost decreased the total organic carbon and carbon-to-nitrogen ratio, as well as increased the pH and microbial population during composting. Interestingly, the pH of compost increased to 5 for the first week of composting, and then it gradually increased to pH around 8 at the end of the process. The results indicated the degradation of organic acids during composting. The neutral to alkaline pH value of compost was reported as an indicator for good aeration of compost pile. The temperature of compost could reach up to 50-55^{o}C during thermophilic phase. At the end of composting, the inoculated compost increased in the total microbial population, especially actinomycetes and total bacteria. The microbial inoculated compost was likely to speed up the composting process of oil palm EFB from 64 days to 50 days.

Keywords: Composting, Microbial additives, empty fruit bunches (EFB).

1. Introduction

Malaysia is the major palm oil production country after Indonesia [1]. The solid wastes from oil palm plantation such as trunks, fronds and oil palm empty fruit bunches (EFB) are a problem for environmental issues. Thus, composting is likely to become an effective alternative in managing the wastes. Technically, composting is a bioconversion process which turns organic wastes into stable amorphous dark brown to black colloidal humus-like substances [2]. Therefore,

Abbreviations

Ctl	Control Sample
EFB	Oil Palm Empty Fruit Bunches
EM	Effective Microorganisms
EMAS	Effective Microorganisms Activated Solution
EMRO	Effective Microorganisms Research Organization
ETC	EM·1™-Treated Compost
FELDA	Federal Land Development Authority
OM	Organic Matter
TOC	Total Organic Carbon Content

composting recycles the organic wastes and the mature compost produced after composting can also be used as bio-fertilizer for a wide range of crops. This microbial added bio-fertilizer is believed to promote soil health for sustainability of crop production.

The addition of microbial additive which is consisted of consortia of beneficiary microorganisms would increase the composting rate and the product quality. One of the commonly used microbial additive in Malaysia is Effective Microorganisms (EM·1™) from Japan. This microbial additive enhances microbial diversity of soil, detoxifies pesticides, suppresses plant disease and soil borne pathogens, as well as enhances nutrient cycling and produces soil enhancing metabolites to increase plant yield [3, 4]. The scientific data of EM·1™ for the type of strains and their composition are often not revealed and generally remained as trade secrets. Therefore, the performance of EM·1™ on EFB was investigated in this study, in term of its physical, chemical and biological properties.

A complete composting process can be examined by various physicochemical and biological parameters including temperature, microbial activities, total organic carbon content (TOC), total nitrogen content, carbon-to-nitrogen (C:N) ratio, formation of humic substances, CO2 production, moisture content and pH level [2, 5]. These parameters are of great importance to be an indicator for compost maturity and stability [6, 7]. The application of immature compost can be problematic to the ecosystem. This is because decomposition process is still continuing and hence inducing the anaerobic conditions in soil pores.

The aim of the present work was to investigate the effects of microbial additive (EM·1™) on the composting of oil palm empty fruit bunches (EFB). The quality of the compost was assessed in terms of their physiochemical composition such as temperature, pH and carbon-to-nitrogen (C:N), and microbial population.

2. Materials and Methods

2.1. Raw materials for composting

The raw materials for composting comprised of oil palm empty fruit bunches (The raw materials for composting comprised of oil palm empty fruit bunches (EFB) collected from FELDA Holding Bhd (Kuala Penggeli, Johor). EFB was ground into small pieces using a grinding machine (CSJ-250, FANQUN®, China) with the particle size of 0.5-20.0 mm. Urea and Molasses were purchased from Merck (Germany) and EMRO (Johor, Malaysia), respectively.

2.2. Preparation of effective microorganisms activated solution (EMAS)

The commercial microbial inoculant product, EM·1™ (Effective Microorganism 1) was purchased from EMRO (Johor, Malaysia). EM was activated by mixing the product with molasses, and water in a composition of 1:1:20. The mixture was stirred, dissolved well and left for 7 days in a glass container without exposure to direct sunlight. It was ready for use when it gave sweet and sour smell after a week. The pH of the EM activated solution (EMAS) was recorded to be around pH 3.5 to 3.7.

2.3. Preparation of composts

Two sets of compost (2kg) were prepared in this study. The first compost was inoculated with EMAS (ETC) and another set of the compost was used as control sample (without EMAS, Ctl). EFB, urea and molasses were used as the key raw materials for composting. Before the process, EFB was sterilized using an autoclave to eliminate the microbes present in EFB. The compost in this study was consisted of 2kg of EFB in 1L of urea (1M) and molasses (400mL). After mixing the raw materials using a garden fork in a 22L container (purchased from JAMOF Sdn. Bhd., Johor, Malaysia) as shown in Fig. 1**,** a 1L of EMAS was added into the ETC and well mixed the compost mixture. However, 1L of water was added into the control samples. Mixing and turning of the compost were conducted thoroughly throughout the composting process in order to ensure good aeration of the compost. The temperatures of the compost were recorded on daily basis throughout the process.

Fig. 1. Compost Bin and Its Dimension.

2.4. Determination of physiochemical properties during composting

The pH of the compost samples were determined by using DELTA 320 pH meter (Mettler Toledo, United State). A water suspension with a ratio of 1:10 of air-dried compost and water was used for the measurement of pH. The organic matter (OM) of the compost was measured by drying in the oven at 103 ^{o}C ± 2 ^{o}C for 6 h, followed by the weight loss after ignition in the furnace with temperature of 560 °C ± 10°C for 7h.Total nitrogen was analyzed using the Kjeldhal method [8].

2.5. Microbiological analysis

EM·1™ activated solution (EMAS) and compost samples were collected for microorganism identification by using spread plate method. For EMAS, the identification was carried out on the fully activated EM, which was the one after a week of activation. For the compost samples, the identification was carried on the 1st day, followed by the 1st, 2nd, 3rd, 4th, 5th and 9th week of composting. In order to identify the mesophiles and thermophiles during different composting phases, the mesophiles was incubated at 30 ± 2°C, whereas thermophiles were incubated at 50 ± 2°C. Four different microorganism groups were identified in this study, namely total bacteria, actinomycetes, lactobacillus and yeast, by using nutrient agar, actinomycete isolation agar, lactobacillus deMan-Rogosa-Sharpe agar and Rose Bengal Chloramphenicol agar, respectively.

3. Results & Discussion

3.1. Physiochemical properties of compost

The temperature profile determines the success of an aerobic composting [9]. Under the optimal conditions, a composting process will go through four phases: (1) First mesophilic phase; temperature ranges from 20 to 40°C, also known as the preparatory stage which initiates the decomposition process; (2) Thermophilic phase that stands for more than three days; temperature rises to 40 to 60°C due to the extensive microbial metabolism; (3) Second mesophilic phase; temperature drops to the mesophilic range which allows the re-establishment of the heat resistant microorganisms; (4) Maturity phase; compost lacks of toxins and detrimental microorganisms with homogenous physical appearance and constant nutrient contents [2, 10].

EFB is highly resistant to microbial attack due to its lignocellulosic properties with an estimation of 44.2% of cellulose, 33.5% of hemicellulose and 20.4% of lignin [11, 12]. Thus, the temperature profile for the composting process was fluctuated in this study with only two phases were observed, which were the thermophilic phase that recorded in the first 10 days of the process and mesophilic phase which last from day 10 to 64. The temperature was increased from 32°C to nearly the thermophilic phase (52°C) for 2-3 hours every day for both microbial inoculated and control samples in the first 10 days. Although the temperature was not maintained in the thermophilic range for long hour, the rise of temperature had proven the high microbial activity in the compost. Throughout the process, microbial inoculated compost gave higher temperature profile than control samples, especially during the first month of the process. This observation explains the active microbial population in microbial inoculated compost resulted from the addition of EMAS. This also indicates the ability of EM·1™ in the degradation of recalcitrant organic matters in EFB and produced the labile carbon sources for other microorganisms.

The pH values during the composting process are listed in Table 1. The starting pH for microbial inoculated compost and control samples were recorded as 4.41 and 4.76, respectively. Lower starting pH was recorded for microbial inoculated compost due to the acidic pH of the EMAS solution which recorded around 3.5-3.7. The pH of the compost gradually increased during composting as a result of organic compound degradation and mineralisation. At the end of the process, the pH reached slightly alkaline value, pH 8.12 and 8.52 for microbial inoculated compost and control samples, respectively. A similar pH trend was also reported by previous investigators in other composting processes using pruning waste, garbage, dairy manure with rice chaff and cow manure [6, 10, 13, 14].

Although both microbial inoculated compost and control sample started with an acidic pH and having an increasing pattern in pH throughout the process, the increment observed in control sample was more alkaline. This rapid increment in pH explains low microbial activities in control sample with low degradation of EFB, and thus the leading to the depletion of labile carbon sources. It is important to highlight that, compost mixtures with high pH values should be avoided since pH more than 7.5 always lead to the loss of N through NH_3 volatization, which always associated with odor problem [15].

For microbial inoculated compost, a more stable pH increment with less alkaline was observed. These results showed that microbial inoculated compost exhibited higher microbial activities than control sample, which also indicated the effectiveness of EM·1™ in degrading the recalcitrant organic matters in EFB. A slightly alkaline pH around 7.5 to 8.5 for the end product of composting could be always obtained [6, 16].

Table 1. pH of Compost during Composting.

Day	pH						
	1	8	15	22	29	36	64
Ctl	4.76	6.50	7.17	7.05	7.69	7.85	8.52
ETC	4.41	5.28	5.97	7.44	7.37	7.76	8.12

Table 2 shows the initial and final reading of total organic carbon (TOC), total nitrogen (TN) and C:N ratio from this study. Although C:N ratio decreased for both microbial inoculated compost and control samples (due to the loss of TOC and the increased in TN content), the composts still did not meet the recommended C:N ratio of 15-20:1, which is an indicator for matured compost [2]. The slight decrease of TOC could be attributed to the slower degradation of the highly fibrous raw material (EFB) and high content of recalcitrant compounds. Similar observation also obtained in composting using sugarcane trash which rich in lignin content [17]. The moderate increase of TN could be attributed to the partial loss of N through the formation of ammonia (especially under high pH and temperature environment). Besides, the raw material which contained relatively high C:N ratio was favoured for microbial N immobilization [17, 18], in which, the organic nitrogen might be degraded to form ammonium or nitrate which were then taken up by microorganisms as nitrogen source for organic compound synthesis. Microbial inoculated compost showed higher decrease in TOC as the decomposition progress, from 52 to 46%. Overall, the final C:N ratio was recorded to be lower in microbial inoculated compost (40.30). This further illustrates the ability of EM·1™ in either EFB degradation or in attracting some cellulase- or hemicellulase-producing strains for EFB digestion.

Table 2. Organic Carbon, Total Nitrogen and C:N Ratio Detected at the Initial and Final of the Composting.

Compost	Organic carbon (%)		Total nitrogen (%)		C:N Ratio	
	Ctl	ETC	Ctl	ETC	Ctl	ETC
Initial	50.23 ± 4.97	52.07 ± 2.95	0.60 ± 0.07	0.67 ± 0.05	53.74	54.15
Final	51.95 ± 2.97	46.06 ± 2.30	0.77 ± 0.08	0.76 ± 0.07	51.74	40.30

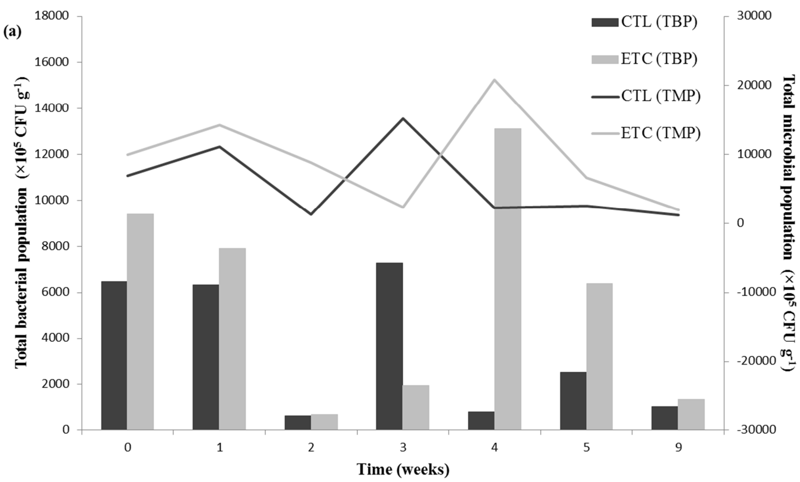
3.2. Microbiological Analysis

In the microbiological analysis of EMAS, the population of mesophilic bacteria, lactobacillus and actinomycetes were found to be 6.20, 13.30 and 81.99 × 10^8 CFU per gram of compost, respectively. However, no mesophilic yeast and fungi, as well as thermophiles were detected in EMAS. The microbial populations during composting were strongly affected by the starting material used. Since EFB was highly fibrous, it contained high yeast and fungi content, therefore the raw materials were autoclaved before composting. Thus, no yeast and fungi was detected in the compost throughout the composting process. The presence of thermophiles started to be detected at the second week of the process, particularly higher population in microbial inoculated compost. This observation explains that microbial inoculant could assist the composting process by attracting microorganisms from the environment. These microorganisms increased the process of composting.

Figure 2 presents the microbial count data collected in compost samples throughout the whole process. In the present study, there was only slight decrease in bacteria population at the end of composting process due to the depletion of nutrient. It is clear that bacteria were able to colonize compost pile at the early stage of the process due to their small size (0.5-3.0 µm) which allowed the rapid transfer of soluble substrates into the cells. It has rapid growth rate on soluble proteins and other readily available substrates. Therefore, bacteria was found to dominate the compost pile throughout the process, with microbial inoculated compost giving higher bacterial population than control sample. The microorganisms took 2 weeks (3^{rd} and 4^{th} week) for environmental adaptation before tremendously increased in their population at the 5^{th} week of composting (Fig. 2(a)). Specifically, actinomycetes were only required a week (4^{th} week) for adaptation as presented in Fig. 2(b).

The population of actinomycetes were observed to maintain around 1.3-18.1×10^7 CFU per gram of compost throughout the process. This result seems reasonable since actinomycetes are common in many environments and able to utilize a wider range of carbon sources and to sporulate prolifically due to their ubiquity. The results obtained in EMAS also showed high population of actinomycetes. Moreover, actinomycetes also secreted a wide range of extracellular enzymes that able to degrade cellulose and lignin, although their lignocellulose degradation characteristics are weaker than fungi, but this difference never reduce their importance in composting process [19]. They are still identified as one of the main groups responsible for organic matter conversion because of their ability to sustain in the harsh environment [19-21]. As highlighted by Goodfellow and Williams [20] and Steger et al. [22], the ideal growth temperature for actinomycetes is within the mesophilic range, some of this species actually can withstand at high temperature during the thermophilic phase, and even become more active when they approach or exceed 60°C.

Although small difference of microorganism population was observed between the inoculated compost and control sample when the composting proceed, control sample required longer time to build up its own microbial population. Thus, the time taken for control sample to reach the maturity phase was about 2 weeks longer than the inoculated compost.



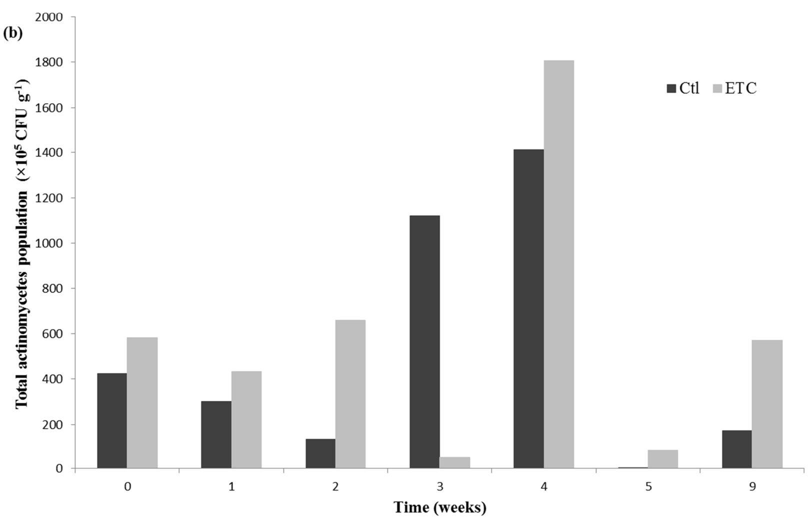


Fig. 2. Microbial Count for (a) Total Bacterial (TBP) and Total Microbial Population (TMP), (b) Total Actinomycetes Population. ETC: EM·1™-Treated Compost (Microbial Inoculated Compost); Ctl: Control Sample.

4. Conclusion

The inoculation of microbial additive into EFB compost can improve the efficiency of EFB decomposition.

- In term of lower total organic carbon content and lower carbon to nitrogen ratio at the end of composting process.
- Further supported by the increase of pH value (~8), microbial population and degradation organic acids as a result of active decomposition.

To further verify the compost quality as a bio-fertilizer, future work on some detailed analysis of other physicochemical and microbiological parameters, as well as the studies on larger scale process are needed.

Acknowledgement

The authors would like to thank Universiti Teknologi Malaysia for the support of the study (04H45) and MOE (Ministry of Education) for the research grant of 04L114.

References

1. Malaysia Palm Oil Board (MPOB) (2014). The oil palm tree. Retrieved June 16, 2014, from http://www.mpoc.org.my/The_Oil_Palm_Tree.aspx.
2. de Bertoldi, M.; Vallini, G.; and Pera, A. (1983). The biology of compost: a review. *Waste Management & Research*, 1(2), 157-176.
3. Monica, S.; Karthik, L.; Mythili, S.; and Sathiavelu A. (2011). Formulation of effective microbial consortia and its application for sewage treatment. *Journal of Microbial Biochemical Technology*, 3(3), 51-55.
4. Higa, T.; and Wididana, G.N. (1991). Changes in the soil microflora induced by effective microorganisms. *Proceedings of the first international conference on Kyusei Nature Farming*. U.S. Department of Agriculture, Washington, D.C., USA.
5. Michael, D.; and Kathleen, S. (2001). Biological, chemical, and physical processes of composting. In Stoffella, P.J.; and Kahn, B.A. *Compost utilization in Horticultural cropping systems*. CRC Press.
6. Benito, M.; Masaguer, A.; Moliner, A.; Arrigo, N.; and Palma, R. (2003). Chemical and microbiological parameters for the characterisation of the stability and maturity of pruning waste compost. *Biology and Fertility of Soils*, 37(3), 184-189.
7. Forster, J.; Zech, W.; and Würdinger, E. (1993). Comparison of chemical and microbiological methods for the characterization of the maturity of composts from contrasting sources. *Biology and Fertility of Soils*, 16(2), 93-99.
8. Bremmer, J.M.; and Mulvaney, C.S. (1982). *Methods of soil analysis, part 2: Chemical and microbiological properties* (2^{nd} E.). ASA, SSSA: Madison, Wis.
9. Hassen, A.; Belguith, K.; Jedidi, N.; Cherif, A.; Cherif, M.; and Boudabous, A. (2001). Microbial characterization during composting of municipal solid waste. *Bioresource Technology*, 80(3), 217-225.
10. Liu, J.; Xu, X.H.; Li, H.T.; and Xu, Y. (2011). Effect of microbiological inocula on chemical and physical properties and microbial community of cow manure compost. *Biomass and Bioenergy*, 35(8), 3433-3439.

11. Taherzadeh, M.J.; and Karimi, K. (2007). Acid-based hydrolysis processes for ethanol from lignocellulosic materials: A review. *BioResources*, 2(3), 472-499.
12. Aziz, A.A.; Das, K.; Mohamad, H.; and Anis, M. (2002). Effects of physical and chemical pre-treatments on xylose and glucose production from oil palm press fibre. *Journal of Oil Palm Research*, 14(2), 10-17.
13. Ishii, K.; Fukui, M.; and Takii, S. (2000). Microbial succession during a composting process as evaluated by denaturing gradient gel electrophoresis analysis. *Journal of Applied Microbiology*, 89(5), 768-777.
14. Liu, D.; Zhang, R.; Wu, H.; Xu, D.; Tang, Z.; Yu, G.; Xu, Z.; and Shen, Q. (2011). Changes in biochemical and microbiological parameters during the period of rapid composting of dairy manure with rice chaff. *Bioresource Technology*, 102(19), 9040-9049.
15. Miller, F.; Macauley, B.; and Harper, E. (1991). Investigation of various gases, pH and redox potential in mushroom composting Phase I stacks. *Australian Journal of Experimental Agriculture*, 31(3), 415-423.
16. Boulter-Bitzer, J.I.; Trevors, J.T.; and Boland, G.J. (2006). A polyphasic approach for assessing maturity and stability in compost intended for suppression of plant pathogens. *Applied Soil Ecology*, 34(1), 65-81.
17. Civeira, G. (2010). Influence of municipal solid waste compost on soil properties and plant reestablisment in peri-urban environments. *Chilean Journal of Agricultural Research*, 70(3), 446-453.
18. Goyal, S.; Dhull, S.K.; and Kapoor, K.K. (2005). Chemical and biological changes during composting of different organic wastes and assessment of compost maturity. *Bioresource Technology*, 96(14), 1584-1591.
19. Sonia, M.T.; Naceur, J.; and Abdennaceur, H. (2011). Studies on the ecology of actinomycetes in an agricultural soil amended with organic residues: I. identification of the dominant groups of Actinomycetales. *World Journal of Microbiology & Biotechnology*, 27(10), 2239-2249.
20. Goodfellow, M.; and Williams, S.T. (1983). Ecology of Actinomycetes. *Annual Review of Microbiology*, 37(1), 189-216.
21. Boulter, J.I.; Boland, G.J.; and Trevors, J.T. (2000). Compost: a study of the development process and end-product potential for suppression of turfgrass disease. *World J Microb Biot*, 16, 115-134.
22. Steger, K.; Jarvis, Å.; Vasara, T.; Romantschuk, M.; and Sundh, I. (2007). Effects of differing temperature management on development of Actinobacteria populations during composting. *Research in Microbiology*, 158(7), 617-624.