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Full Length Research Paper

Composting of food and yard wastes by locally isolated fungal strains

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Four lignocellulolytic fungi isolates, *Phanerochaete chrysosporium* PC-13 (PC2094), *Lentinus tigrinus* M609RQY, *Aspergillus niger and Penicillium* spp. were used as inocula in source separated organics (food and yard trimmings) from solid waste (SW) to produce biofertilizer and stabilize waste constituents. The results show that composting with sequential pure culture accelerates substrates decomposition and reduce bulk volume. The initial moisture content was 64%, and the percentage C/N ratio ranged between 8.60 and 18.20%, while the produced compost C/N ratio was <25%, indicating its viability for large scale production since the acceptable C/N ratio for efficient compost is >12%. The 74% total organic matter (TOM), 7.2 pH and 132% germination index (GI) further showed the potentials of the produced compost. Based on this, food waste (FW) and yard trimmings (YT) showed an economic potential for sustainable production of compost using low technology.

Key words: Lignocellulolytic, composting, fungi strain and biofertilizer.

INTRODUCTION

Municipal solid waste (MSW) generation increases with the rise in population and change in lifestyle. Statistically, Asia produced the largest amount of Urban Food Waste (UFW), which is expected to increase from 251 to 418 million tonnes (45 to 53% of total world UFW) from 1995 to 2025 (Adhikari et al., 2009). Currently, the 17000 tonnes of waste generated per day in Kuala Lumpur comprises of 57% food wastes, 17% mixed papers, 4.7% yard trimmings and others constitute the MSW generated and disposed (MH and LG, 2008; Nasir, 2007; Saeed et al., 2009). However, Malaysia waste treatment data revealed that 50% of these wastes are openly dumped, 30% are land filled, 5% are incinerated and only 10% are composted (Nasir, 2007). This open dump of organically rich wastes contributes significantly to the formation of leachate quality and quantity aside from the spread of disease vectors, odour, aesthetics and other environmental damages. Leachates constitute major threat to underground water and the eco-system due to the presence of heavy metals. This situation prompted the continual pressure of the Malaysia local authorities in seeking appropriate strategies to deal with the MSW menace (Saeed et al., 2009).

Currently, the conventional methods of waste management is accredited with problems such as open dump of comingled wastes which contribute significantly to the formation of primary leachate quality and quantity which are threat to surface and underground waters (Opatokun et al., 2010). Bye-products of incineration such as volatile organic compound (VOC), fly ash and heavy metals constitute both health and environmental threats (Franchini et al., 2004). Meanwhile, the present demand for biofertilizer had increased due to its environmental friendliness as compared to synthetic fertilizers which always have adverse effects on humans and animals.

Therefore, the interception of organic wastes at source for the production of biofertilizer will not only solve the

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Abbreviations: SW, Solid waste; **TOM,** total organic matter; **GI,** germination index; **FW,** food waste; **YT,** yard trimmings.

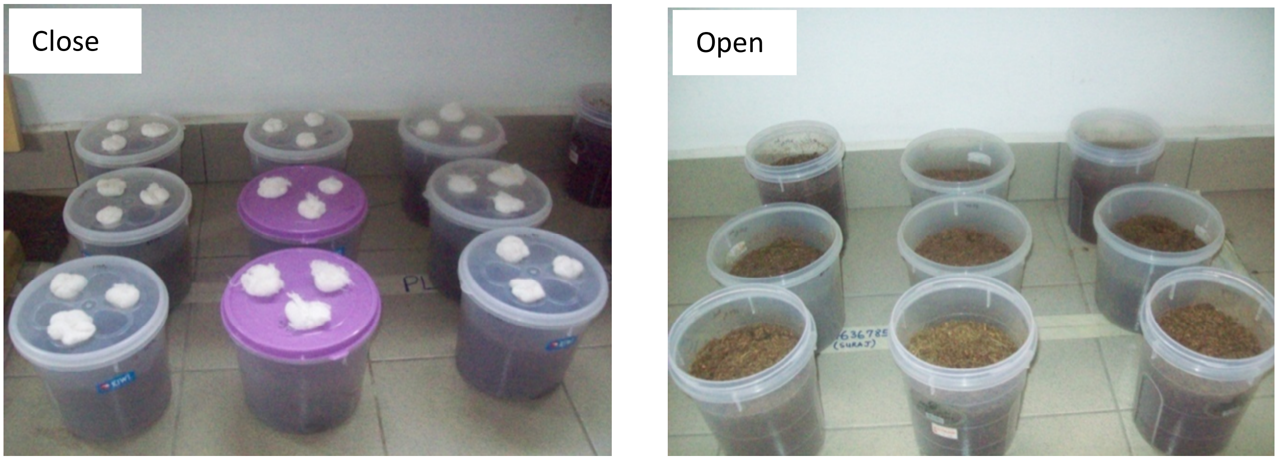

Figure 1. Close and open systems.

health and environmental problems constituted by these ‘resources out of place’ but increase their economic value. This recycling process stabilizes the wastes, increases the life span of landfills, eliminate VOC formation during incineration and reduce the greenhouse gases (GHG) generated in landfills by providing a more sustainable and environmental friendly treatment technique.

The aim of this study was to provide a sustainable method that enhances decomposition of food wastes and yard trimmings using locally isolated lignocellulolytic fungi strains in the production of a biofertilizer and/or soil amendment.

MATERIALS AND METHODS

Four lignocellulolytic fungi isolates were used in the current study. These strains were isolated from different sources and locations in Malaysia. From previous study, the potential for lignin degradation has been shown by both *Phanerochaete chrysosporium* PC-13 (PC2094) and Lentinus tigrinus M609RQY (Tijani et al., 2011), while *Aspergillus niger* indicated decomposition of cellulose and hemicellulose (Alam, 2009) and, *Penicillium* spp. shows ability to degrade fibers and disinfect (Molla et al., 2004). These selected strains of *P. chrysosporium* PC-13 (PC2094), *L. tigrinus* M609RQY (IMI 398363), *A. niger* and *Penicillium* spp. were from the Department of Biotechnology Engineering, International Islamic University Malaysia (IIUM) laboratory stock. They were all maintained on 3.7% w/v potato dextrose agar (PDA) plate except L. tigrinus which was maintained on 4.8% w/v of malt extract agar (MEA). The young peripheral edges of the culture were used for sub culturing after cutting with a cork borer (d = 5 mm). All the fungi were incubated for 5 days at 32℃, except L. tigrinus that was incubated for 6 days.

Four sets of plastic bowls/pots, containing 400 g of substrates mixtures were prepared for the composting experiment. All substrates were autoclaved at 121℃ for 50 min after the addition of nutrients. Two sets of system, open (WRo) and close (WRc) were carried out. Each set consist of triplicate plastic pots. The two main substrates, food waste (FW) and yard trimmings (YT) were mixed along with sawdust (SD) for solid state bioconversion (SSB) process. The ratio of 1:1:0.5 (w/w) was used to mix FW (69.10% moisture, 48.85% TOC and 3.18% TKN), YT (69.88% moisture, 50.82% TOC and 1.46% TKN), and SD (19.32% moisture, 53.39% TOC and 0.27% TKN). The substrate to water ratio of 30:70% was used to maintain the optimum moisture content that is peculiar to SSB within the range of 50 and 60% (Haug, 1993; Sherman, 2005). Nutrients (gkg-1) of K_2HPO_4 (0.3), NaCl (0.3) and $MgSO_4.7H_2O$ (0.3) were added to substrate as starter dose for activation of inoculated (*P. chrysosporium*) fungi. Sawdust was also added as a cheap source of carbon (Adhikari et al., 2008).

Thereafter, the containers were inoculated with 6% fungal spores/mycelia out of the entire 70% moisture constituent (59% distilled water, 6% inoculum and 5% minerals) of each opened and closed system. The inoculum sizes used were 2.5 x 107 and 5.5 x 107 spores per ml for *P. chrysosporium* and L. tigrinus, while 84 x 106 and 92 x 106 CFU/g air dried inoculum for *A. niger* and Penicillium spp., respectively. Composting plastic pots were kept at room temperature with the open system uncovered and holes were created on the lids of the closed system which were covered with cotton wool (Figure 1). The main characteristics of the mixture were: pH = 5.68, TKN = 2.50%, TOC = 51.15%, EC = 5.62 mS/dm (Wb), salinity = 5.62 0/00 (Wb) and TDS = 2.77 g/l. Likewise, the hemicellulose, cellulose, lignin and water soluble contents were 32.30%, 21.60%, 16.00% and 30.10, respectively (Table 1).

The SSB process was evaluated with 10 harvests or samplings (0, 5, 10, 15, 20, 30, 40, 45, 50 and 60 days, respectively) by determining the moisture content, total organic carbon (TOC) content, total Kjeldahl nitrogen (TKN) contents, pH, ash content, degree of degradation (DD) and germination index (GI). Results were analyzed using one-way ANOVA.

RESULTS AND DISCUSSION

The average moisture content of 64% obtained is acceptable (Sherman, 2005) even though the preferred range is 50 to 60% depending on the substrate and the fermentation process adopted. In the closed system (WRc), beside the ambiguous drop in the value of the control sample, waste reactor with inoculum turned out to be relatively higher. This can be traced to the covering which disallowed the evaporation process to take place within the system. Meanwhile, the water holding capacity of *P. chrysosporium* (PC) and *A. niger* (ASP) was higher in this system as compared to *L. triginus* (LT) and *A.*

Table 1. Characterization of substrates.

Substrates	FW	YT	SD	Mixture
pH	5.39	5.97	4.60	5.68
*TKN	3.18	1.46	0.27	2.50
*TOC	48.85	50.82	53.39	51.15
EC (mS/dm) w.t	3.04	4.13	0.320	5.62
Salinity (0/00) w.t	1.6	1.8	0.1	2.77
TDS (g/l) w.t	3.06	2.06	0.151	2.72
*Hemicellulose (%)	43.37	31.90	18.07	32.30
*Cellulose (%)	11.30	25.73	30.50	21.60
*Lignin (%)	1.87	21.53	45.93	16.00
**WSC (%)	43.46	20.84	5.50	30.10

FW = Food waste, YT = Yard trimmings, SD = Sawdust; *Dry weight basis, wet basis (w.t), ** water soluble components (WSC). TOC, Total organic carbon; TKN, total kjeldahl nitrogen; EC, electrical conducitvity.

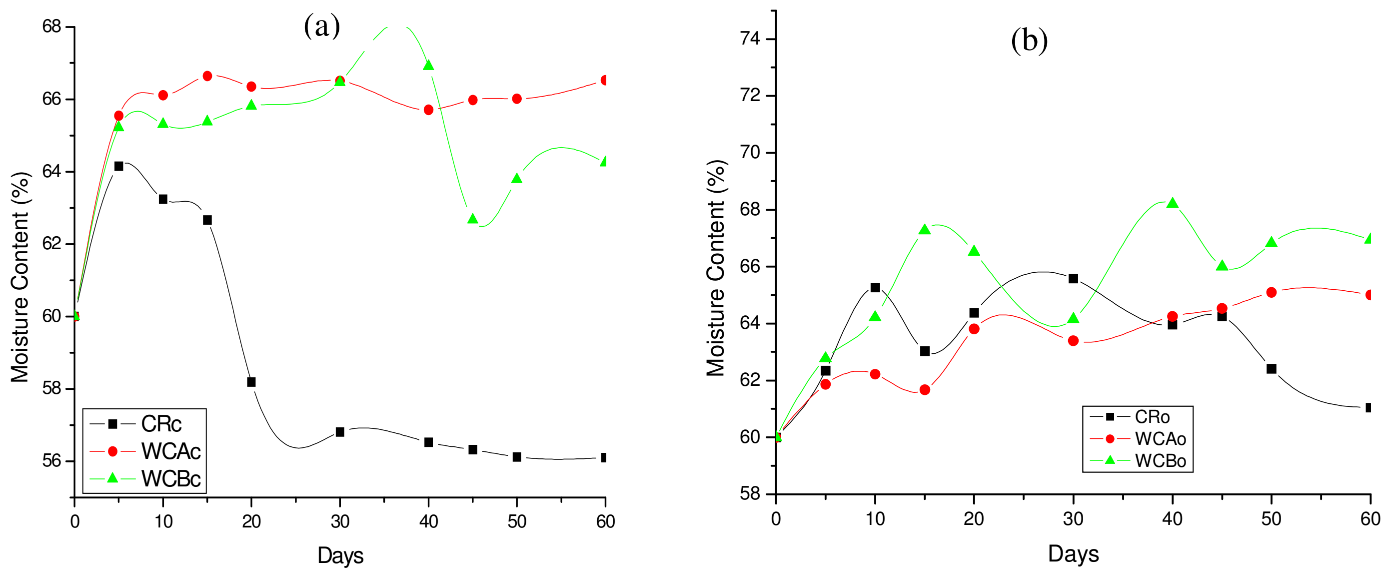

Figure 2. Moisture content of (a) close and (b) open systems.

niger. Subsequently, after the inoculation of *Penicillium* spp. (PEN) the moisture content of WCBc increased, then decreased at a greater rate, thereafter stabilized at day 60 (Figure 2a and b). Statistically, the open and close system are significant ($P < 0.05$), meanwhile, the system with *P. chrysosporium* (WCAc) and *L. triginus* (WCBc) indicates a least square difference (LSD) value of 0.001 with respect to the control (CRc), while *P. chrysosporium* (WCAo) and *L. triginus* (WCBo) are slightly significant with $P = 0.047$. This implied that at 95% confidence, it can be proved that the moisture content of the close system is significantly different.

Total organic carbon (TOC) shows a significant correlation with time by maintaining a positive correlation with the TOM (Figure 3a and b). The decreasing trend of TOC over time had been widely considered as a measure of composting performance indicator (Figure 3c and d). Sampedro et al. (2007) reported that total carbon losses in solid substrate cultures are usually associated with a decrease in OM due to mineralization which is also directly related to respiration. In this study, the open system showed a statistically significant difference of 0.022 in the mean value of control (CRo) against WCAo and WCBo. Meanwhile, the LSD in post-hoc was used to further determine where the difference in the mean lies for CRo with respect to WCAo and WCBo which relates a significant value of $P = 0.015$ and $P = 0.017$ for WCAo and WCBo, respectively compared to CRo. Moreover, the homogeneity value of 0. 901 indicates that the results (WCAo and WCBo) had not violated the homogeneity of variance assumption. An equivalent one-way analysis of variance was conducted on the closed system to explore the difference in the CRc, WCAc and WCBc.

There was a statistically significant difference at P ≤

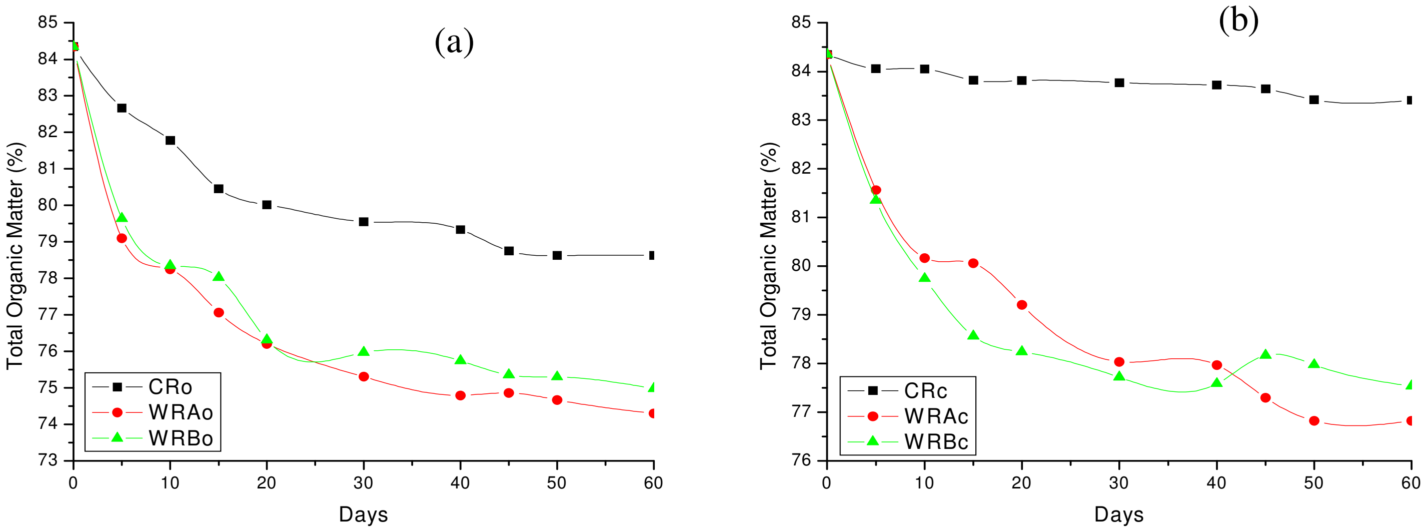

Figure 3. Total organic matter (%) of (a) open and (b) close systems and total organic carbon (%) of (c) close and (d) open systems.

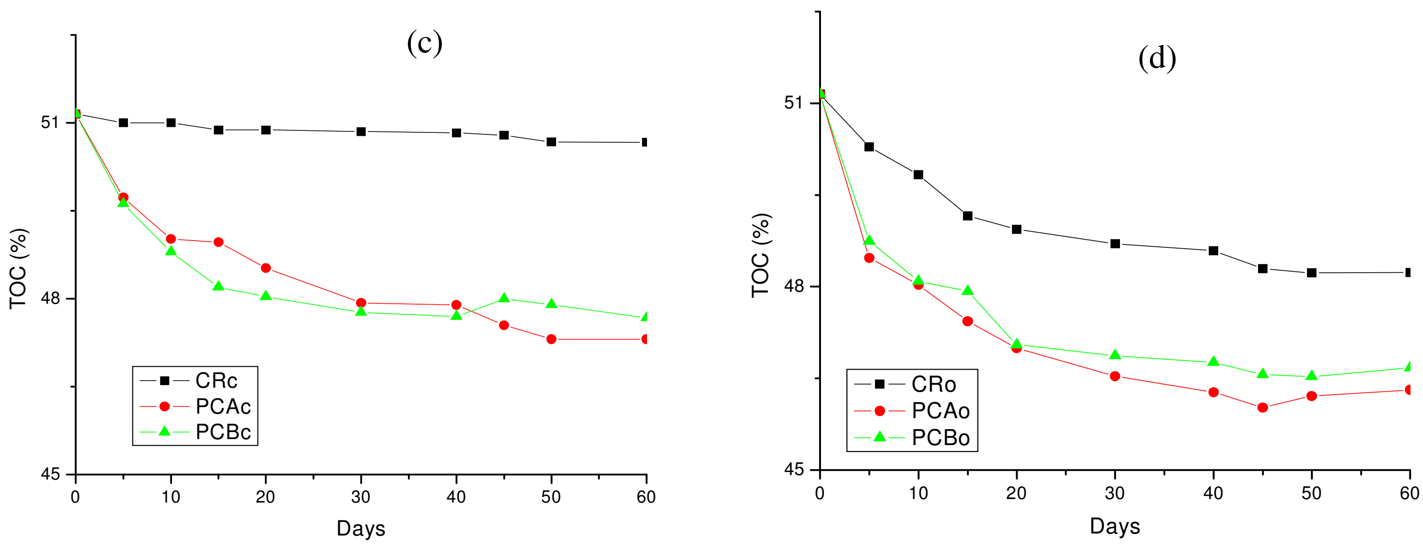

Figure 3. Contd.

0.05 in TOC value for the system [F (2, 26) – 19.4, P = 0.001]. Despite being statistically significant, the actual difference in the mean values between WCAc and WCBc was quite small. Post-hoc comparisons using LSD test indicates that the mean differences for *P. chrysosporium* and *L. triganus* are also significant. Nitrogen increases across the two systems (open and closed). The trend is relatively unstable within each of the systems (Figure 4a and b) with few oscillations between the initial value of 2.50% and the maximum of 3.09% in open, while in closed it ranges between 2.46 and 2.89%. The increase in TKN due to the concentration effect caused by the degradation of the labile organic carbon contents reduces the weight of the composting mass. The TKN concentration usually increases during the composting process when the loss of volatile solid (organic matter) exceeds the loss of NH_3 (Lee, et al., 2007). The increasing trend of TKN could be accredited to the addition of starter salts (K_2HPO_4, $MgSO_4.7H_2O$ and NaCl) which has been reported to be responsible for the reduction in nitrogen loss (Eklind and Kirchmann, 2000; Jeong and Kim, 2001). The subsequent decrease in the TKN especially on irregular basis can be linked to the high frequency of turning which hastened volatilization (Ogunwande et al., 2008). Statistically, the closed system is significantly different at $P \le 0.05$ level in TKN value for the system [F(2, 26) = 8.5, P = 0.001]. Meanwhile, the LSD post-hoc values of CRc with respect to WCAc and WCBc indicate

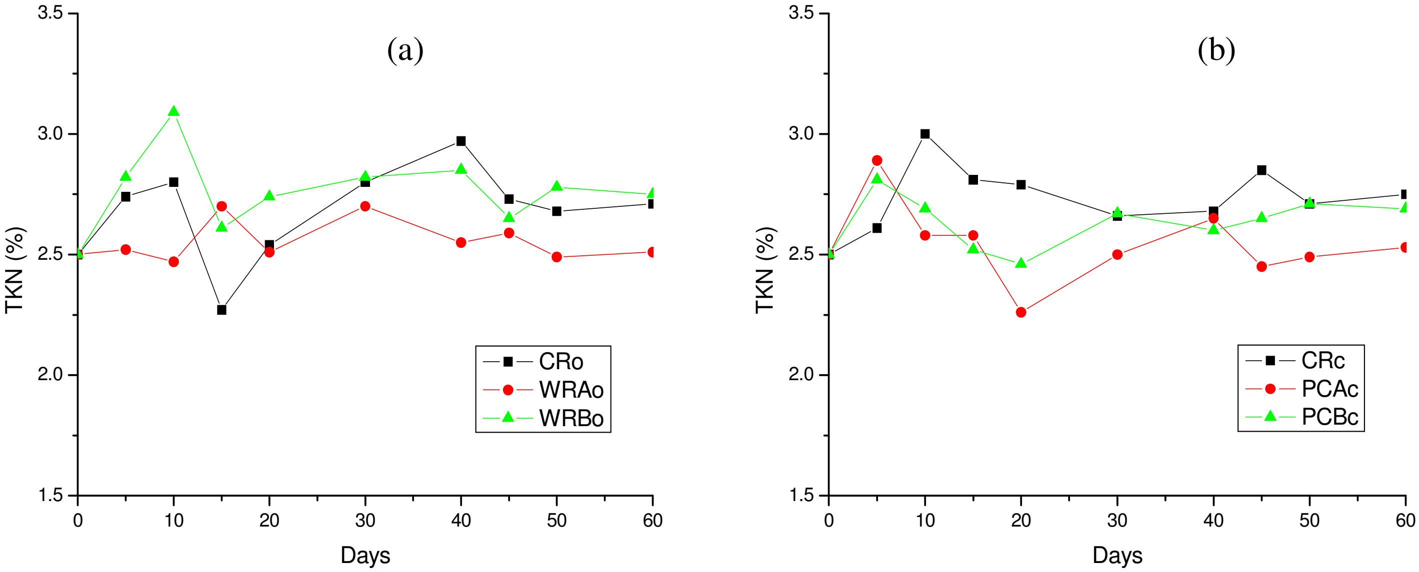

Figure 4. Total Kjeldahl nitrogen (%) for (a) open and (b) close systems.

a significant difference only in WCAc (0.009), while WCBc (0.216) is not significant in relation to CRc.

Generally, the slower rate of C/N decomposition may be linked to the relative high lignin content of the substrate mixture (16%) and other organic acids formed (Haddadin et al., 2009) during the composting process. The open system shows (5.23 to 7.60%) a proportional decrease across the control (CRo) and the treated samples in WCAo and WCBo. Since carbon constitutes most of the plant biomass accumulated through photo-synthesis, carbon losses can be expected during FW and YT composting because the principle is to reduce the bulk and increase the nutrient content of the substrate mixture. The substrate mixture with 43.37% (Table 1) of lower chain carbon (hemicellulose) can easily decom-pose during composting through microbial mineralization of the organic matter.

This is followed by oxidation of carbon during the SSB process to CO2, part of which was released to the atmosphere, and the other part may dissolve to form the weak carbonic acid. The declining pattern of TOC and the oscillating values of TKN provide a balance condition which is essential for microbial activity (Benito et al., 2003). Meanwhile, the closed system shows a significant difference across the one-way ANOVA [F (2, 26) = 10.9, P = 0.001] at $P \leq 0.05$ level, but the LSD indicates that only WCBc is significant (P = 0.001), while WCAc is not (P = 0.154). The open system is not significantly different (P = 0.454) in both the LSD and $P \geq 0.05$ as shown in ANOVA for both WCAo (P = 0.530) and WCBo (P = 0.213), respectively.

The minimum pH recorded was about 5.68 at the beginning of composting. After 10 days of composting in the open system, there was a rapid and significant ($P \leq 0.05$) increase in pH to 8.82, but it declined thereafter as composting progressed. This increase could be attributed to the degradation of organic acids and the production of ammonium ions (Haddadin et al., 2009). Beck-Friis (2000) also reported that fatty acids present in household waste were only partially degraded in externally heated compost, thus as the pH and NH3 emissions increases, the fatty acid degradation occurs. The loose nature of the open system (control inclusive) as regards the possibility of microbial population and subsequently microbial activity could be responsible for such peaks of pH in the system.

However, the pH trend in the closed system was between the range of 5.68 (initial) and 7.3. This could be due to the presence of organic acids in the leachate generated, low microbial activity and sensitivity of the microbes to the acidic condition of the system. Moreover, the existence of one microbial community in the waste, a mesophilic acid-tolerant (*P. chrysosporium* and *L. triganus*) ensures the initial degradation of organic contents (Sundberg et al., 2004).

Germination index which combines the measure of relative seed germination and relative root elongation of seed is used to evaluate the toxicity and degree of maturity of the compost (Zucconi et al., 1981). GI of tomato seeds generally decreases gradually and signifi-cantly reaches the minimum at days 7 and 15 in the open and days 10 and 15 in the closed system, respectively (Figure 5a and b). This decrease might be due to the release of high concentration of ammonia and low molecular weight of organic acid (Fang et al., 1999). The minimum value of GI in the open system was 53 and 55%, while in the closed system it was 49 and 56%, respectively.

In all situations, the values of GI increased to about 90% and above which correspond to the suggestion of

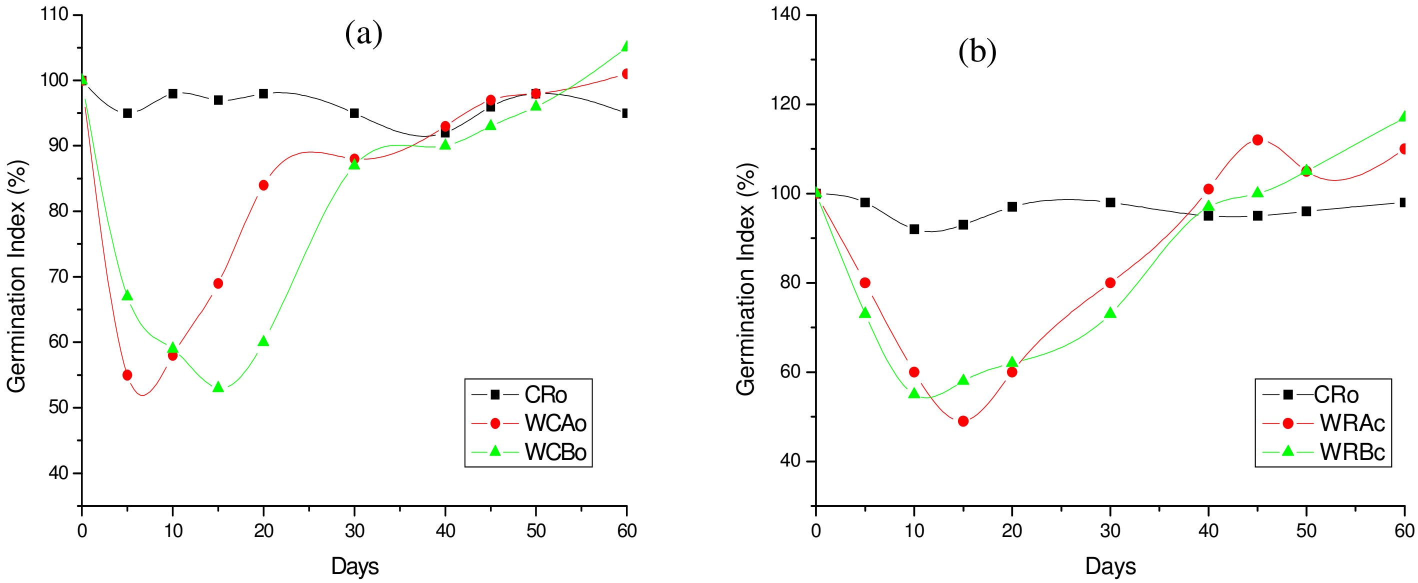

Figure 5. Germination index (%) for (a) open and (b) close systems.

Jimenez and Garcia (1989) that seed GI value above 50 is suitable for agriculture utilization, while GI value above 80% indicates that the compost is mature (Tiquia, 2003; Zucconi et al., 1981).

The significant GI increase at day 15 through 40 during the SSB process could be due to relief phytotoxins, especially the ammonia volatilization, reduction of unstable organic acids and probably the anti microbial strength of Aspergilus and Penicillium spp. (Gaind et al., 2009). Several species of Aspergillus and Penicillium may coproduce ochratoxin A and penicillic acid when cultured on organic substrates (Batista et al., 2003). Meanwhile, at harvest, the highest level of GI was 105 and 132 in open and closed system, respectively. GI trend as indicated shows further increasing effect even beyond the 60 days harvest period.

Generally, the open system is significantly different at $P \leq 0.05$ level with value [$F (2, 27) = 3.478$, $P = 0.045$] with post–hoc LSD values 0.047 and 0.023 for WCAo and WCBo, respectively. While, the closed system is not significant ($P \geq 0.05$) with value [$F (2, 27) = 1.305$, $P = 0.288$] and the post–hoc LSD of 0.376 and 0.120 for WCAc and WCBc when compared with CRc. The microbial effect was determined based on the significant percentage decrease of C/N ratios 5.37, 5.23 and 7.60% in the WCAo stream coupled with 6.68, 4.73 and 5.58% in WCBo stream on days 15, 40 and 60, respectively in the open system. Similarly, 2.32, 1.74 and 4.54% of the WCAc stream and 5.01, 5.32 and 3.05% of WCBc for days 15, 40 and 60 of the close system shows that LT activity is higher in the close system than PC. These could be responsible for the higher activity shown by *A. niger* (ASP) in day 40 coupled with the mineralization potential of ASP and PEN as active cellulose and hemicelluloses degraders.

Moreover, the DD is higher in open system (11.92%) as compared to closed system (8.93%) which indicates that the effect of other microbes in the SSB process is almost not significant. Similarly, the significance of the SSB reaction order indicates R2 values of 0.984 and 0.981 for open systems, while closed systems were 0.865 and 0.965 for WRA and WRB, respectively.

L. tigrinus performed actively at pH slightly below normal (5.68 to 6.37) in the closed system, while PC performed at a lower pH of 5.45 to 6.03 during 15 days SSB process of the duo. PC result is in agreement with other studies (Alam et al., 2002; Haddadin et al., 2009; Molla et al., 2004) which show its optimal output in an acidic medium. PC performance was generally better than that of LT with respect to the degree of degradation; the later had average DD values of 6.86%, while PC recorded 8.64% as the highest in both organisms, especially at day 15 which is the expected expiration time of the duo activities. This performance of PC could be responsible for its wide adoption (Alam et al., 2001; Fakhru'l-Razi et al., 2002) as an environmentally friendly SSB organism.

However, between the closed systems, LT performance was relatively better than PC, which suggests why LT had the lowest OM, TOC and C/N values in the closed system at day 15. Consequently, the germination index of the open systems were considerably low as compared to those of the closed system, this could be as a result of the higher microbial activities in the open system as compared to the close system where intrusion of microbes are restricted. The toxicity strength as expressed by the GI indicated that open system (53 to 55%) produced a significantly toxic biomass as compared to the close system (49 to 56%), especially during the most active degradation period (day 15). Similarly, TOM is significant (P = 0.001) in both open and close system, while C/N ratio of the closed system is significant only between systems and the open is not significant at $P \leq 0.05$.

Conclusion

Generally, PC behaviour in the open system is better than the LT of the closed. The values of C/N ratios 5.37, 5.23 and 7.60% in the WCAo stream coupled with 6.68, 4.73 and 5.58% in WCBo stream on days 15, 40 and 60, respectively indicate the microbial effect in the open system. Similarly, 2.32, 1.74 and 4.54% of the WCAc stream and 5.01, 5.32 and 3.05% of WCBc for days 15, 40 and 60 of the close system show that LT activity is higher in the close system than PC. These could also be responsible for the higher activity shown by ASP in day 40 coupled with the mineralization potential of ASP and PEN as active cellulose and hemicellulose degraders. Finally, there is need for cost benefit analysis of these two systems, especially before commercialization or large scale production. Moreover, some of the parameters need to be further optimized for precise values. These techniques thus proved sustainable for most developing countries as it require the use of SSB with a low technology concept.

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