Mineralization of Poly(lactic acid) (PLA), Poly(3-hydroxybutyrate-co-valerate) (PHBV) and PLA/PHBV Blend in Compost and Soil Environments

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ABSTRACT: The present study investigates the mineralization of poly(lactic acid) (PLA) and poly(3-hydroxybutyrate-co-valerate) (PHBV), and PLA/PHBV blend in compost and soil burial environments. The mineralization was assayed on the basis of carbon dioxide (CO_2) release from the test materials incubated in compost and soil for a period of 200 days. The degradation was followed by means of fragmentation, thermogravimetric (TGA), FTIR spectroscopy and scanning electron microscopy (SEM) analyses. The results showed that PLA, PHBV and blend of PLA/PHBV achieved almost 90% biodegradation under composting conditions, while PHBV, PLA/PHBV blend and PLA respectively achieved only 35%, 32% and 4% biodegradation under soil burial conditions. The results of crystallinity, thermal degradation, microstructure and surface morphology analyzed by TGA, FTIR and SEM indicated that the blending approach and different environmental conditions in soil and compost were the most influencing parameters for the ultimate biodegradation of these biobased polymers.

KEYWORDS: Biodegradation, PLA, PHBV, PLA/PHBV blend, compost and soil burial

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1 INTRODUCTION

Nowadays, biobased polymers are emerging as alternatives to replace petroleum-based synthetic polymers which pose severe environmental pollution problems in terrestrial and aquatic habitats [1–3]. Most commonly used synthetic polymers are derived from nonrenewable resources and they are very resistant to biological degradation (biodegradation) and therefore persist in the environment for many years [2, 3]. This has led to increased research interest over the last two decades for the development of biobased and biodegradable materials. This research has been aimed at the best practices in waste management based on sustainable uses of carbon sources through mechanical, chemical and biological recycling. Biobased polymers are synthesized using the monomers derived from renewable resources. They have shown remarkable improvements in their properties, which have increased their usage as eco-friendly materials in various industrial sectors such as automotive, pharmaceutical, medical, packaging, consumer products and others [4]. They are also widely utilized for short-term and single-use applications in packaging, trash bags, mulch films, personal products and some home care products [4]. The market share for biobased polymers is growing rapidly [1, 4]. According to an analysis conducted on European bioplastic products and their demand [5], the worldwide production capacity for biobased polymers is predicted to increase from approximately 1.4 million tons in 2015 to 6 million tons in 2019, which accounts for an annual growth rate of 32.7% during this five year period [5]. Although biobased polymers are more expensive than traditional polymers, in recent years new technological development in the production of biobased polymers from non-food agricultural biomass and lignocellulosic waste have resulted in a substantial decrease in their cost. The market share of biodegradable polymers in the biobased plastics market is currently about

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90%. Forecast suggests that in 2016 large production capacities of bioplastics will be established; 46.3% in Asia and 45.1% in South America, followed by 4.9% in Europe and 3.5% in North America [4].

Biobased polymers from renewable resources, including those produced by chemical synthesis of biobased monomers, such as poly lactic acid (PLA), and those produced by microorganisms, such as poly(hydroxybutyrate-co-valerate) (PHBV), show properties comparable to traditional polymers [6, 7]. Both PLA and PHBV have received considerable attention from the research community due to their biocompatibility and thermoplasticity. However, these biobased polymers have some limitations due to thermal resistance, lower mechanical properties and environmental performance, which restrict their diverse applications. Therefore, several approaches to modify and improve their properties by means of physical-chemical, thermal properties and environmental performances in abiotic and biotic conditions have been studied. Indeed, the blend preparation of PLA with other bioplastics like polyhydroxylalkanoates (PHA) or biodegradable polymers like polycaprolactone (PCL) and polybutylene succinate (PBS), is considered as a common approach for tailoring their properties without losing the biobased attributes and biodegradability of the final products [8, 9]. Further studies to understand the influence of physical-chemical, thermal and mechanical properties of biobased polymers for evaluating the ultimate biodegradability at different natural environments is needed. A comprehensive study about degradation and biodegradation mechanisms of these polymers is necessary for their safe use in various end-use applications. International norms and standards are available for testing biodegradability of polymeric materials in natural environment, which are key prerequisites with respect to both environmental and food safety aspects. This approach has generated much research interest in exploring processability and evaluation of the properties of the polymer blends for potential applications.

It is well know that PLA degrades by hydrolysis reactions of the ester bond. Hydrolysis degradation of PLA has been extensively studied by several researchers [10–12] and it has been found that the degradation is influenced by size and geometry of the sample [13], degree of crystallinity [14], molecular weight [14], isomer ratio [13] and environmental conditions such as temperature, pH and microorganisms [15–18]. Moreover, plasticizers such as citrate esters, glycerol or their copolymer are often used to incorporate into PLA-based materials in order to improve inherent brittle properties, thermal stability and durability during their shelf life. It is shown that addition of plasticizers helps in exchanging the intermolecular bonds between macromolecules and low molecular compounds, thus enabling conformational changes and resulting in improved thermal, mechanical and gas barrier properties [19–21]. Extensive review of the literature shows that PLA is one of the most studied biobased polymers among other renewable polymers. The degradation of PLA is well reported under simulated composting conditions (58–60 °C), unlike real environments such as soil, anaerobic digester and marine water [15]. Some highlighted results showed that a significant weight loss in PLA was observed after one year soil burial [22, 23]; however, since the weight loss is not directly related to biodegradation, complete mineralization tests for monitoring CO_2 and CH_4 evolution are needed. PHBV, the other bacterial biopolymer used in this study, is also well known for enzyme-catalyzed hydrolysis and the functional groups undergo biodegradation in soil and compost environments. Naoyuki and Doi [24] and others [25–26] have studied the hydrolysis and biodegradation behaviors of PHA homopolymers and copolymers P3HB with varying concentrations of hydroxy valerate (HV) content. They reported that the degradation rate of PHA was several times higher than that of PHB and PHBV and they found that the enzymatic hydrolysis reaction with PHA degrading enzymes have more influence for hydrolytic degradation rather than abiotic hydrolysis degradation in water [26]. Also, the rate of degradation was mainly affected by degree of crystallinity and lamellar structures of PHB and PHBV [26].

Recently, many research studies on degradation of PLA, PHBV and their modified blends were mainly studied by weight reduction, loss of mechanical properties and morphology variations under different environmental conditions; however, they do not provide sufficient evidence on the final biodegradation step of polymeric carbon conversion into CO_2 as well as its physical-chemical degradation process of the fragmented residues. In this article, the degree and rate of ultimate biodegradation of PLA, PHBV and PLA/PHBV blend in soil and simulated compost conditions were monitored. The changes in physical-chemical, thermal and morphological properties of their fragmented residues during biodegradation were also evaluated by using DSC, TGA, FTIR and SEM techniques.

2 EXPERIMENTAL SECTION

2.1 Materials

Commercially available PLA of injection molding grade, 1001(CP-INJ-1001-EZC), with a density of 1.28 g.cm^{-3} and melt flow index of 8 g/10 min at 190 °C, was obtained from Cereplast, Inc. (Hawthorne, NY, USA) in pellet form which contains additive derived from cereal starch (wheat, potato and tapioca). Commercially available PHBV biopolymer of injection molding grade, BV326301 (PHB88/PHV12), with a density of 1.25 g.cm^{-3} and melt flow index of 15 g/10 min at 190 °C, was obtained from Goodfellow Cambridge Ltd. (UK) in pellet form, which was plasticized with 10% citric acid. All other chemicals used in this study were of analytical grades and were used as received from Sigma Aldrich, South Africa.

2.2 Methods

2.2.1 Preparation and Processing of Polymers and Blend

The PLA/PHBV (70/30 wt%) blend was developed by melt compounding process in a co-rotating twin-screw extruder, with an length/diameter ratio of 40 (CTE-20, Coperion, China) equipped, with a feeder and a strand pelletizer. The PLA and PHBV pellets were dried in a convection oven at 50 °C for 8 hours before extrusion. The temperature profile during extrusion was set between 180 to 195 °C along the barrel. The screw speed was maintained at 45 rpm for all experimental runs. After exiting the die, the extrudate was solidified by quenching in a water bath before pelletization. The compounded pellets were dried in an oven at 50 °C for 8 hours. For biodegradability testing of PLA, PHBV and PLA/PHBV (70/30) blend, a solution casting method was employed to prepare polymer films (average 10–11 micron thickness) by dissolving 5 g of polymer in 25 ml chloroform at room temperature in a round glass Petri dish of 35 mm × 10 mm in dimensions.

2.2.2 Biodegradation Studies

2.2.2.1 Biodegradation under Composting Conditions

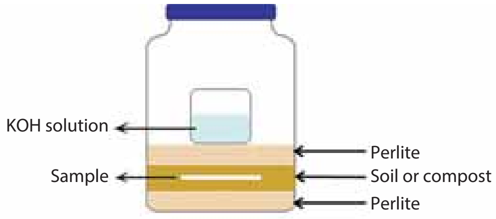
The compost biodegradation of PLA, PHBV and PLA/PHBV blend were conducted under controlled composting conditions, which simulated industrial composting environments. The incubation conditions for the simulated compost biodegradation test were partially adopted from ASTM D5338-15 standard [27]. The biodegradation test was performed in 1000 ml biometric flasks and the test procedures were followed as suggested by Chiellini *et al.* [28] and Stloukal *et al.* [14]. In this biometric test method, compost samples (3 months old and well aerated) derived from a mushroom composting farm were used as incubation medium. A standard protocol was followed to characterize total dry solids, volatile solids, pH and C/N ratio of the compost according to ASTM D5338-15 standard. Table 1 shows the physical and chemical properties of compost samples used in this study. Polymer test samples, compost inoculum and perlite components were weighed and placed into the biometric flasks. 15 g of perlite wetted with 15 ml of distilled water was placed at the bottom layer of flasks. Test materials mixed with compost in a ratio of 1:6 (w/w sample to dry solids of compost) were placed on top of the perlite. Thereafter, another 15 g of perlite wetted with 15 ml of distilled water was placed on the compost layer (Figure 1). Finally, a glass beaker containing 50 ml of 0.5 M KOH solution was placed on top of the upper layer to trap carbon dioxide emission during the biodegradation process. Cellulose (reference substrate) and blank (compost) sample were also included for comparison. The sample flasks were incubated in an air ventilated oven at a temperature of 58 ± 2 °C, which simulated industrial composting conditions. Three replicates for each sample were tested. At 1–3 day intervals, the CO_2 absorbed in the KOH solution was back titrated with 0.1 N HCl by adding 1 N $BaCl_2$ solution before the titration for absorbing alkaline medium. After titrations, each beaker was washed and refilled with fresh standard solution of 0.5 M KOH. The activity of compost inoculum with reference to CO_2 emission showed that 59.2 mg CO_2/g volatile solids were produced in the first 10 days, which is within an acceptable range as prescribed for compost studies [27]. The moisture content of the test flasks were recorded with a moisture analyzer. The moisture content of each flask was maintained at 50–55% throughout the test by measuring the weight loss due to evaporation, and adding deionized water to counterbalance such losses.

Table 1 Physical and chemical properties of the compost and soil material.

Analysis	Compost	Soil
Total dry solids (%)[a]	55	80.9
Volatile solids (%)[b]	53	23
pH of compost solution	7.1	7.2
Total organic carbon amount (%)	10.6	3.4
Total nitrogen amount (%)	0.9	0.12
Carbon/nitrogen ratio (C/N)	11.8	28.3

[a] The amount of solids obtained by taking a known volume of compost and drying at about 105 °C for 10h.

[b] The amount of solids obtained by subtracting the residue of a known volume of compost after incineration at about 550 °C.



(**a**) Schematic diagram of biometer flask

(**b**) A biometer flask used in this study

(**c**) Biometer flasks in oven at 58 ± 2 °C for compost biodegradation studies

Figure 1 A real biometer flask respirometric system used for studying the biodegradation of polymeric materials.

2.2.2.2 Biodegradation under Soil Burial Conditions

The soil biodegradation of PLA, PHBV and PLA/PHBV blend were performed under laboratory room temperature of 23–25 °C under dark conditions to simulate the real soil environment. The biodegradation test conditions and the procedures were followed as per the ASTM D5988-12 standard [29]. The soil for biodegradation tests was obtained from Grassmaster Inc., Port Elizabeth, South Africa. Fresh soil from the surface layer of an agricultural field was obtained. Table 1 shows the physical and chemical properties of the soil used. Soil burial biodegradation tests were carried out in a biometer flask of 1 liter capacity (Figure 1). Soil samples sieved at 0.6 mm and supplemented with 10 ml of 0.1% $(NH_4)_2HPO_4$ solution, were used to bury the polymer test samples as shown in Figure 1. The test materials, PLA, PHBV, PLA/PHBV blend; and reference (cellulose) were placed in the middle layer of soil at approximately 16 mg/g sample to dry soil ratio (Figure 1). All test materials and blank were run in triplicate. Test cultures and blank were incubated in the dark at room temperature. In order to measure the CO_2 production from the samples under investigation, including blank, each test vessel was equipped with a beaker containing 40 ml of 0.1 N KOH solution which was substituted every 1–3 days and back titrated with 0.1 N HCl.

2.2.2.3 Calculation

The total carbon dioxide, CO_2(t), emitted by organic metabolization of the material through the action of microorganisms is representative of the total degradation due to biological factors. The total organic carbon of the polymer samples (C_t) was determined by elemental analysis. Equation 1 below was used to calculate the theoretical CO_2 in the total dry plastic material.

$$\mathrm{CO_2}(t) = M_t \times C_t \times \frac{44}{12} \tag{1}$$

where M_t is the total dry weight of constituents or plastic material added to the compost or soil, and C_t is the relative weight of total organic carbon in the total dry plastic material. The degree of biodegradation for each test material was calculated as a percentage of the overall theoretical CO_2 (Equation 2).

$$\text{Biodegradation}(\%) = (\mathrm{CO_2})s - \frac{(\mathrm{CO_2})c}{(\mathrm{CO_2})t} \times 100 \tag{2}$$

where $(CO_2)s$ and $(CO_2)c$ are the amounts of CO_2 produced in the sample and in the control (blank), respectively.

2.3 Characterization

To study the thermal, structural and morphological characteristics of the biobased polymers and their blends at various stages of biodegradation, a known amount of fragmented particles were carefully removed and evaluated to determine the effect of mineralization on the polymers. The test sample removed from the compost or soil was thoroughly washed with deionized water and dried in an oven at 40 °C for 4 h and then characterized by DSC, TGA, FTIR and SEM.

2.3.1 *Differential Scanning Calorimetry (DSC)*

Using a Diamond DSC PerkinElmer instrument, DSC analysis was carried out under nitrogen atmosphere and the following conditions were maintained: first heating from 25 to 200 °C at a rate of 10 °C/min, followed by cooling from 200 to –50 °C at a rate of 5 °C/min and heated again from –50 to 200 °C at a rate of 10 °C/min. The first heating results were used to remove the thermal history of the samples. The regions of melting temperature (Tm) and percentage of crystallinity (Xc) were determined from the second heating scan.

2.3.2 *Thermogravimetric Analysis (TGA)*

A thermogravimetric analyzer (PerkinElmer Pyris) was used to analyze the thermal degradation behavior of the samples by heating from 30 to 600 °C at a rate of 20 °C/min under nitrogen atmosphere.

2.3.3 *Fourier Transform Infrared Spectroscopy (FTIR)*

The FTIR analysis was performed using a Perkin Elmer Spectrum 100 at ambient temperature of 23 °C. The analysis of the samples was done in ATR spectra mode within a wavelength range of 4000–400 cm^{-1} at a resolution of 4 cm^{-1} and averaged over 32 scans.

2.3.4 *Scanning Electron Microscopy (SEM)*

A FEI Quanta 200 (Eindhoven, The Netherlands) scanning electron microscope was used at an accelerating voltage of 20 kV in low vacuum to observe the surface morphology of test material before and after biodegradation. A gold palladium coating of 20 nm thickness was coated on the surface of test specimens to avoid electron buildup during analysis.

3 RESULTS AND DISCUSSION

In order to suit polymers for various practical applications the lifetime of a polymer can be controlled by additives such as starches, antioxidants, coloring agents, sensitizers and plasticizers or chemicals. This additives may significantly alter the biodegradability of the polymer matrix and its products. The degradation mechanism of polymers containing hetero-chain backbone, such as PLA and PHBV, by chemical degradation occurs via hydrolysis or enzyme-catalyzed hydrolysis. The hydrolytic biodegradation mainly depends on the hydrolytic enzymes secreted by microorganisms, which influence the degradation of the long molecular chains of polymers into low molecular weight oligomers and monomers. Based on these facts, standard biodegradation tests monitor the amount of CO_2 released from finished polymeric products. Measurement of released CO_2 and comparison with the theoretical amount based on the carbon content in the original plastic sample provide direct evidence of the extent and rate of biodegradation [28, 30]. The fact that CO_2 emissions are measured during the test proves that the material has entered the natural carbon cycle instead of just deteriorating into biostable microscopic parts which are hard to detect, but would persist in the environment over an unpredictably long period of time. Moreover, the validity of test methods, such as composting and soil burial biodegradation procedures can be confirmed by testing cellulose (positive reference), which is known to be readily biodegradable material. Our results showed that 70% of cellulose biodegraded within 45 days under composting conditions (Figure 2), whereas equivalent biodegradation was prolonged up to 110 days in the case of soil burial tests. These results confirmed that the composting and soil burial methods adopted in this study are suitable for testing biodegradation of polymeric materials as prescribed in ASTM and ISO standards.

3.1 Compost Biodegradation

Figure 2 shows the percentage biodegradability of PLA, PHBV and PLA/PHBV blend carried out in a biometer flask for incubation period of 200 days under controlled composting conditions. The achieved

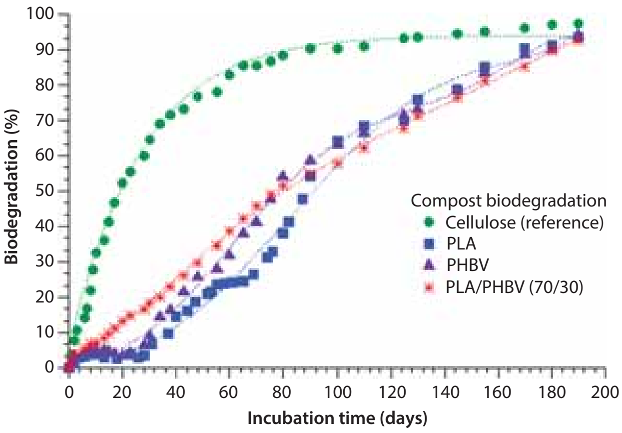


Figure 2 Compost biodegradation results of PLA, PHBV and PLA/PHBV blend.

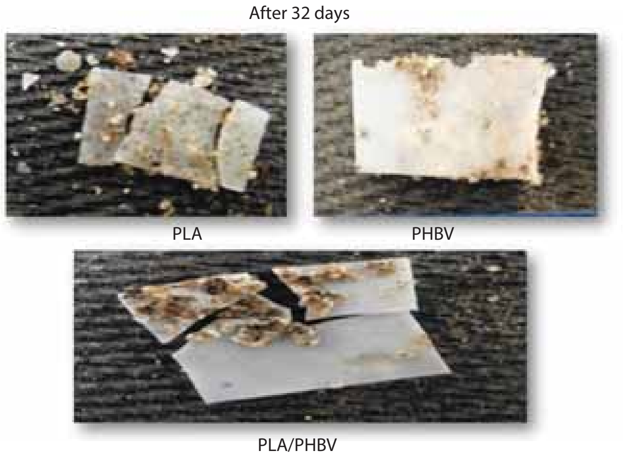


Figure 3 Photographs of PLA, PHBV and PLA/PHBV blend after 32 days of compost degradation.

biodegradability results of these materials under composting conditions were greatly influenced by environmental conditions such as pH (7–8), temperature (58–60 °C) and presence of microorganisms. The results showed that no appreciable amount of CO_2 emission was observed for PLA during the first 32 days of compost incubation, however, some fragmentation was visible, as shown in Figure 3. After the initial phase, which reached 70% biodegradation within 120 days, a slight stationary phase reaching 92% biodegradation in 200 days was observed. This observed degree of biodegradation for PLA in this study is completely different from that reported in other studies showing 60% mineralization of PLA within 60 days [31]. This delayed enzymatic hydrolysis and slow biodegradation could be due to the crystalline and brittle nature of the PLA grade used in this study. On the other hand, the biodegradation of PHBV polymer in compost followed a similar trend to PLA, where the delays in threshold embrittlement behavior started after 32 days. The delayed degradation for PHBV could be attributed to about 12% valeric acid content in the PHBV polymer. Other studies [32] on the biodegradation of PHB and PHBV containing different concentrations of valeric acid were studied under controlled composting conditions and the results showed that PHB was more susceptible to disintegration than PHBV. This is because the presence of 3-hydroxy valerate groups modify the physical, chemical and brittle behaviors of PHBV. However, as shown in Figure 2, after 32 days, the CO_2 emissions of PHBV showed an acceleration phase reaching 70% biodegradation in 100 days and 90% in 200 days, with relatively short stationary phase. It is interesting to note that the observed results for PLA/PHBV blend showed almost 20% biodegradability in the first 32 days when no significant degradation was observed in both PLA and PHBV. This behavior could be attributed to the blending process of polymers, which helps in breaking the crystalline structure of PLA, as well as the surface-enriched heterogeneous phase in one component [33].

During this composting test, samples of PLA, PHBV and PLA/PHBV blend started to undergo fragmentation/disintegration in their respective flasks after 32 days incubation (Figure 3). It was also visually noticed that the fragmented samples became very tiny and not distinguishable in the composting medium after 45 days. The physical degradation of the biopolymers observed after 1 month compost biodegradation clearly indicated that the adopted environmental conditions were suitable for primary degradation resulting in fragmentation/disintegration. Moreover, the characterization of these fragmented residues by physical-chemical, thermal and morphology analyses may provide sufficient technical information for elucidating the degradation mechanisms of these biopolymers.

Figure 4 and Table 2 show the DSC results of PLA, PHBV and PLA/PHBV blend before and after biodegradation. The results showed major changes in the thermograms (melting temperature, crystallinity and heat of fusion) of PLA, PHBV and PLA/PHBV blend after 1 month compost biodegradation. From the DSC thermogram, two melting temperatures, a minor peak and a major peak, were observed for PLA, PHBV and PLA/PHBV blend (Figure 4); and this melting behavior for PLA may be due to PLA containing 10% starch additives and 12% hydroxy valerate (HV) in the PHBV copolymer plastized with 10% citric acid. After 1 month compost biodegradation, the melting temperatures (Tm) of PLA and PHBV and the corresponding percent crystallinity (Xc) were significantly decreased. On the other hand, after 1 month of compost biodegradation of PLA/PHBV blend, the first melting peak corresponding to PLA completely disappeared (Figure 4c and Table 2). These results indicated that PLA and PLA/PHBV (70/30) were rapidly degraded under the adopted controlled composting conditions as compared to neat PHBV. Also, it was visually noticed that after 42 days of compost biodegradation tests all the samples were not distinguishable in the compost medium.

Figure 5 and Table 2 show the TGA results of PLA, PHBV and PLA/PHBV (70/30) blend before and after biodegradation. A decrease in onset temperatures for PLA, PHBV and PLA/PHBV blend after 30 days incubation in compost was observed in comparison to that for neat PLA, PHBV and PLA/PHBV blend (Figure 5). The analyses of the PLA and PHBV fragments after 30 days showed the onset temperature of 221.7 °C and 240.6 °C respectively, whereas those of neat PLA and neat PHBV were 298.7 °C and 253.7 °C respectively. This accounts for the weight loss of

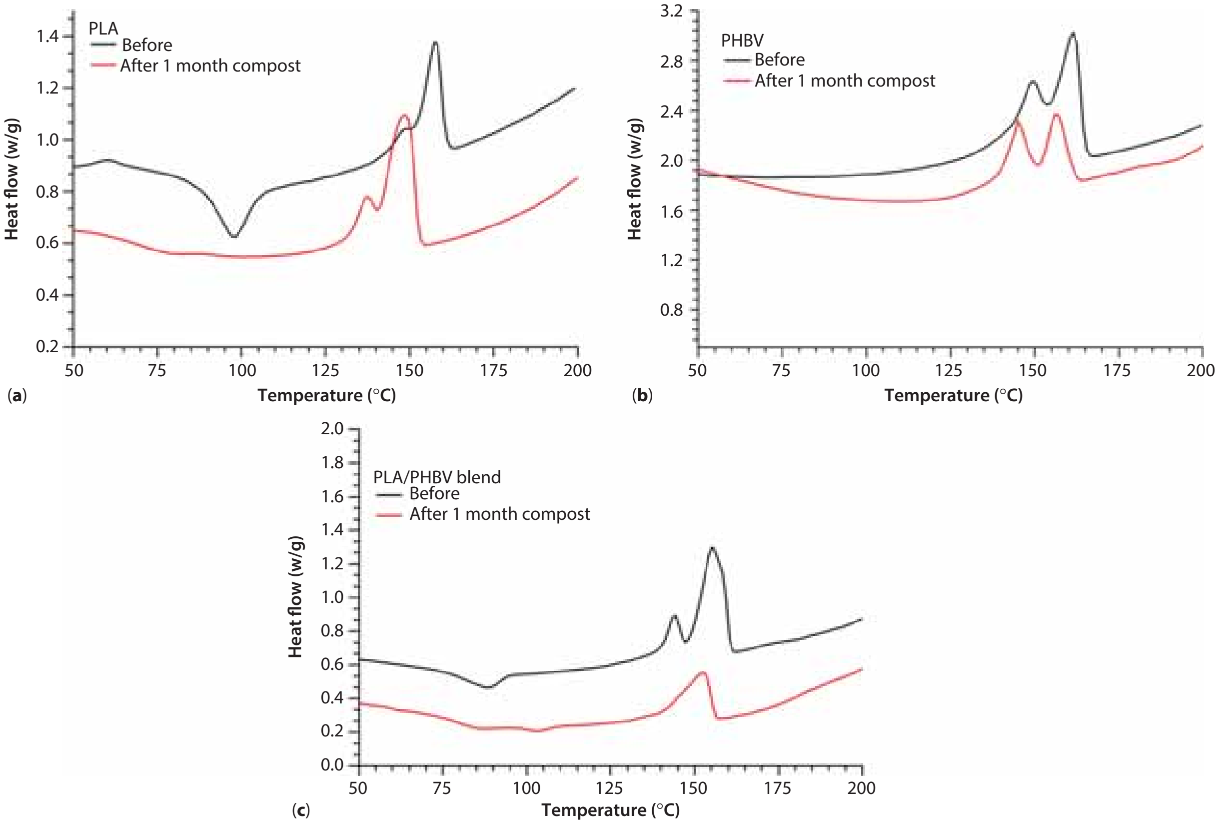


Figure 4 DSC analysis of (**a**) PLA, (**b**) PHBV and (**c**) PLA/PHBV (70/30) blend before and after 1 month of biodegradation under compost conditions.

Table 2 DSC and TGA results of PLA, PHBV and their blend (PLA/PHBV) before and after compost and soil burial biodegradation.

			DSC						TGA		
Sample	**Incubation**	**Days**	**ΔH_m (J/g)**	**ΔH_c (J/g)**	**Tc (°C)**	**Tm_1 (°C)**	**Tm_2 (°C)**	**^{a}Xc (%)**	**T onset (°C)**	**Max T (°C)**	**Residue at 600 °C (%)**
PLA	None	0	19.5	9.7	97.7	148.6	157.7	20.8	298.7	363.8	10.1
	Soil	30	10.4	3.7	99.3	145.4	153.5	11.1	213.2	291.7	18.5
		200	9.8	3.6	99.7	145.4	153.5	10.5	226.1	314.6	25.9
	Compost	30	8.6	14.5	89.8	137.1	148.6	9.2	221.7	329.2	5.9
		200	n.d	n.d	n.d	n.d	n.d	n.d	n.d	n.d	n.d
PHBV	None	0	77.0	85.9	104.0	154.6	162.8	72.6	253.7	285.6	1.5
	Soil	30	78.8	43.0	92.7	145.4	156.5	74.3	242.3	275.9	7.3
		200	28.2	59.9	81.3	146.0	153.6	26.6	213.6	247.5	8.8
	Compost	30	31.8	45.6	94.3	149.4	160.1	30.0	240.6	274.8	1.6
		200	n.d	n.d	n.d	n.d	n.d	n.d	n.d	n.d	n.d
PLA-PHBV blend	None	0	31.8	12.7	71.2	143.9	155.6	23.8	258.7	297.7; 360.0	6.0
	Soil	30	19.0	4.7	95.2	142.4	154.3	14.2	169.05	200.0; 267.4	5.2
		200	13.6	no peak	no peak	no peak	154.1	10.2	195.8	224.5; 298.9	2.1
	Compost	30	7.8	no peak	no peak	no peak	152.1	n.d	224.5	260.5; 300.9	25.1
		200	n.d	n.d	n.d	n.d	n.d	n.d	n.d	n.d	n.d

n.d – not detectable visually in the compost medium

The percentage of crystallinity (Xc %) was calculated from the DSC analysis results: $\Delta H_m/\Delta H_m^0 \times 100$. ΔH_m is the enthalpy of fusion and enthalpy of crystallization. ΔH_f^0 is the enthalpy of fusion for PLA (93.6 J/g) or PHBV (109 J/g) 100% crystalline in each case [34–36].

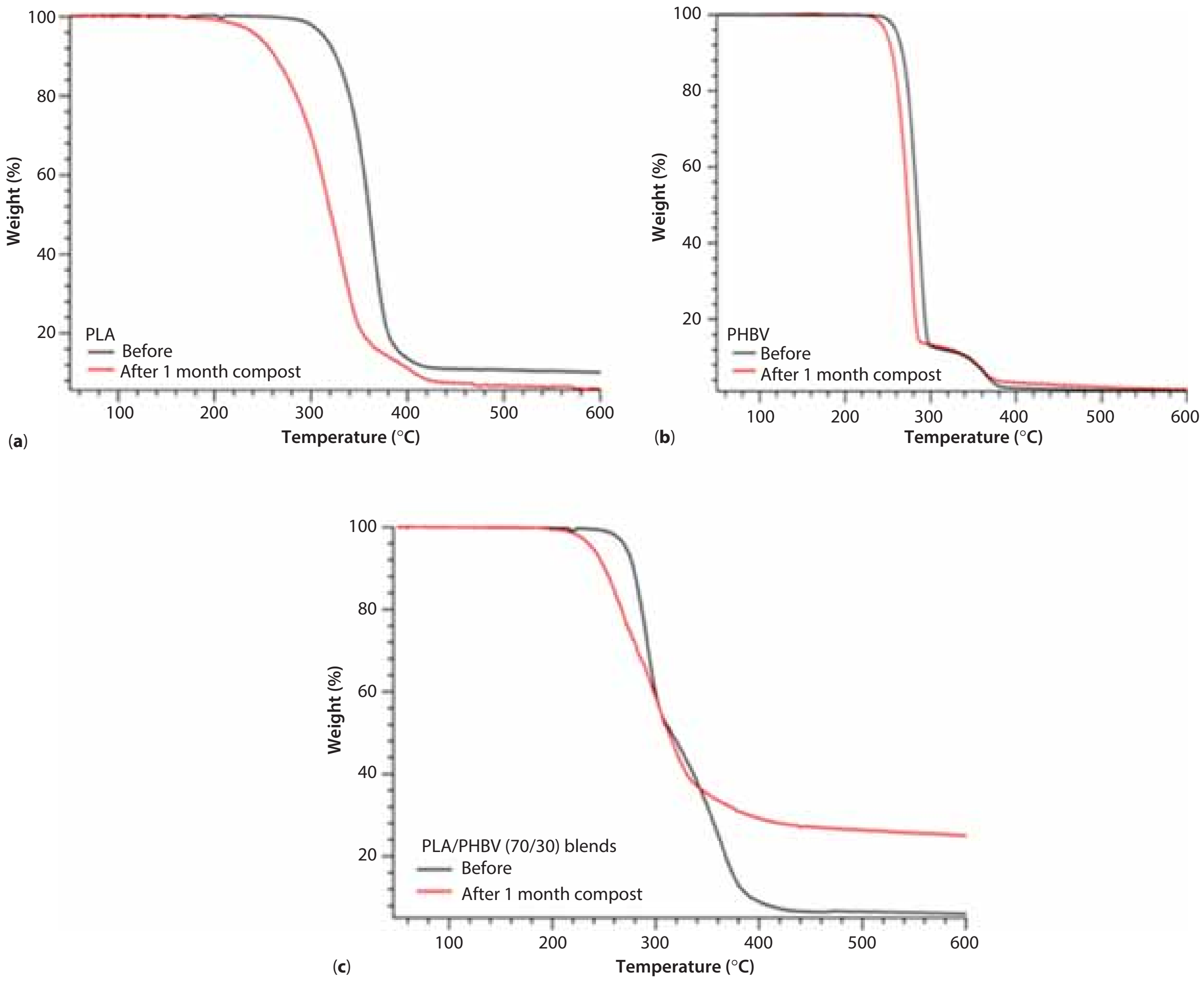


Figure 5 TGA analysis of (**a**) PLA, (**b**) PHBV and (**c**) PLA/PHBV (70/30) blend before and after 1 month of biodegradation under compost conditions.

25.7% and 5.16% for PLA and PHBV, respectively. The maximum weight loss of PLA and PHBV for composted sample occurred between the temperature ranges of 250–350 °C and 253.7–300 °C, respectively, in comparison to that of neat PLA and neat PHBV of 350–380 °C and 240.6–285.6 °C, respectively (Figure 5). The TGA results of PHBV showed two-step degradation behavior with a minor degradation in the temperature range of 300–370 °C, which could be due to 10% citric acid added to PHBV as plasticizer during formulation (Figure 5). The TGA results of PHBV after composting show that there was no significant change in the temperature of minor degradation peak at 360 °C. This result suggests that the presence of microorganisms in the adopted compost conditions were able to degrade the material at 274.8 °C rather than the component whose degradation temperature corresponds to 360 °C. In the case of PLA/PHBV (70/30) blend, it may be interesting to notice that the blend contains some miscibility where the two major degradation peaks occurred at 297.7 °C and 360.0 °C, which correspond to PHBV and PLA, respectively (Table 2). However, the blend does not show any minor peak corresponding to PHBV (Figure 5). These results suggest that during melt processing of the PLA/PHBV blend, the minor degradation peak of PHBV in the temperature range of 300–380 °C could have disappeared due to some compatibility between PLA and PHBV. During the compost incubation of PLA/PHBV blend after 30 days, the weight loss occurred at the onset temperature of 224.5 °C in comparison to that of PLA/PHBV blend at 258.7 °C, which account for 13.2% weight loss after biodegradation (Figure 5). The major degradation peaks of PLA/PHBV blend corresponding to PLA and PHBV after 30 days in compost have shown weight loss of 16% and 12%, respectively (Table 2). These analyses clearly showed that blending of PLA/PHBV accelerated the biodegradation process in comparison to the neat polymers.

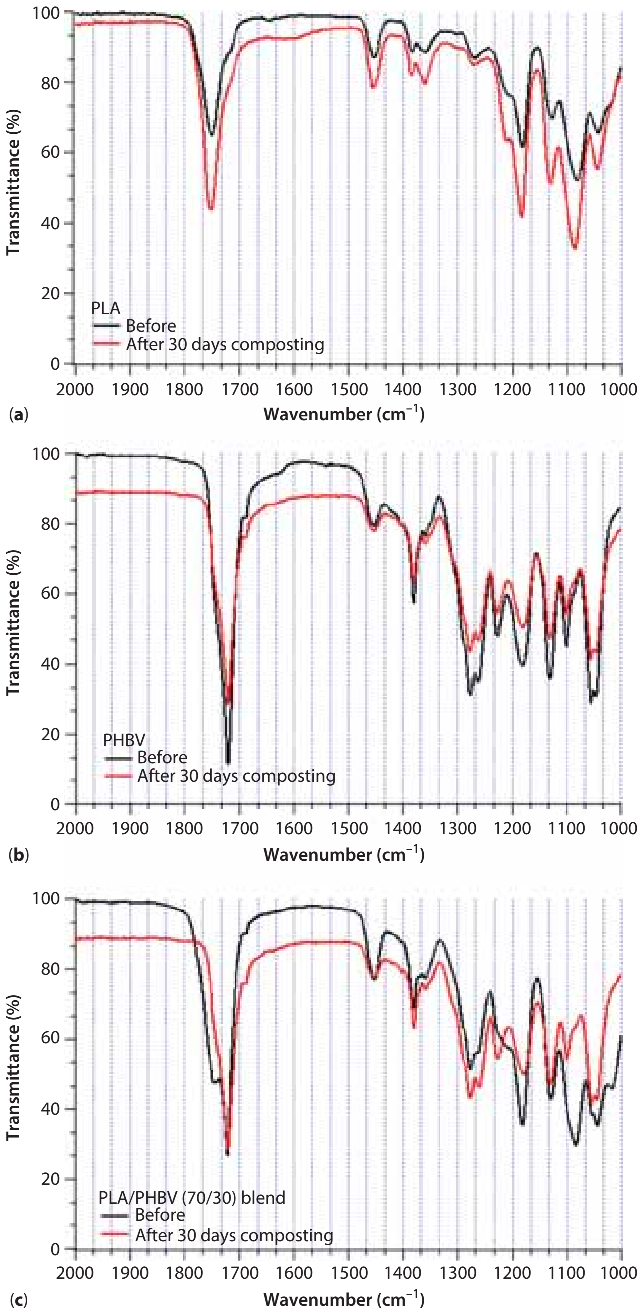


Figure 6 FTIR characterization of PLA, PHBV and PLA/PHBV blend before and after biodegradation for 30 days under compost conditions.

Figure 6 shows the FTIR spectra of (a) PLA, (b) PHBV and (c) PLA/PHBV before and after biodegradation in compost for 30 days. The FTIR results of PLA showed broadened intensity of carbonyl groups (1780–1700 cm^{-1}) and increased peak at a wave number of 1750 cm^{-1} after 30 days of compost biodegradation (Figure 6a). The broadening of carboxylic groups in PLA is mainly due to the hydrolytic degradation process mediated by compost incubation conditions. In the case of PHBV polymer, intensity of carbonyl groups (1775–1680 cm^{-1}) was sharpened and reduced after 30 days of compost biodegradation. These results indicate a loss of carboxylic groups from PHBV. In the case of PLA/PHBV blend, the intensity of broadened carbonyl groups (1780–1680 cm^{-1}) peak at wave numbers of 1750 cm^{-1} (PLA) and 1725 cm^{-1} (PHBV) were observed. After compost biodegradation of PLA/PHBV blend for 30 days, the peak corresponding to a wavenumber of 1750 cm^{-1} completely disappeared and the peak corresponding to a wavenumber of 1725 cm^{-1} was sharpened and reduced in intensity. These results indicate that during compost biodegradation the hydrolytic degradation of carbonyl ester and carboxylic acid components are being assimilated by microbes. Hydrolytic chain scissions at multiple locations in the heterochain polymer produce smaller molecules or oligomers, which can easily permeate out of the polymer matrix. Therefore, reduction in the carbonyl absorbance at a wavenumber of 1710 cm^{-1} results from the enzymatic hydrolysis process (Figure 6) [37, 38]. During the mineralization process, these low molecular weight functional compounds, such as ketone (1715 cm^{-1}), aldehyde (1710 cm^{-1}) and ester (1715 cm^{-1}), are water soluble and they are easily assimilated by microbes. Moreover, microorganisms, such as bacteria and fungi, have a substantial influence on both the rate and overall biodegradation of PLA, PHBV and PLA/PHBV blend in compost environment. It is generally accepted that hydrolysis is the initial process of biodegradation of PLA and PHBV, which is influenced by different enzymatic specificity of microorganism under composting environments [39, 40].

Figure 7 shows SEM photomicrographs of the test samples before and after biodegradation for 30 days under composting conditions. The surface morphology of the samples reveals that the surface degradation of PLA/PHBV blend is more susceptible to undergo biodegradation when compared to neat PLA and PHBV, as shown in Figure 7. The results indicate that it is noteworthy to conduct a detailed investigation on the enzymatic pattern and microorganism population for PLA and PHBV for rendering a clear explanation of the biodegradation mechanisms of biobased polymers.

3.2 Soil Biodegradation

The biodegradation profiles of PLA, PHBV and PLA/PHBV (70/30) blend by soil burial method are shown in Figure 8. The CO_2 emissions recorded in the soil cultures mixed with PHBV and PLA/PHBV blend were significantly higher than those produced from the blank runs, thus indicating the propensity to biodegradation

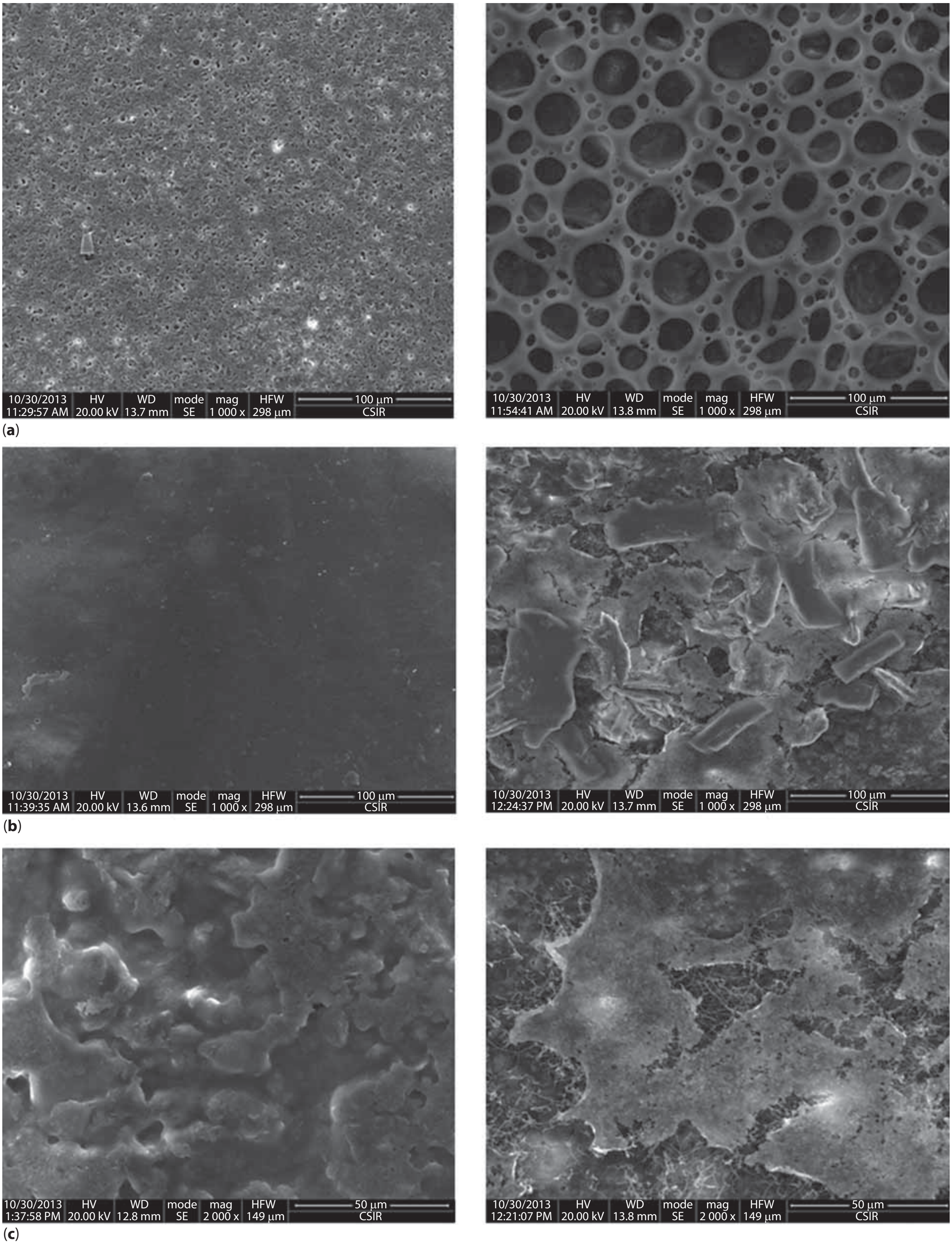


Figure 7 SEM micrographs of (**a**) PLA; (**b**) PHBV; (**c**) PLA/PHBV (70/30 blends) before (left side) and after 1 month of biodegradation under compost conditions (right side).

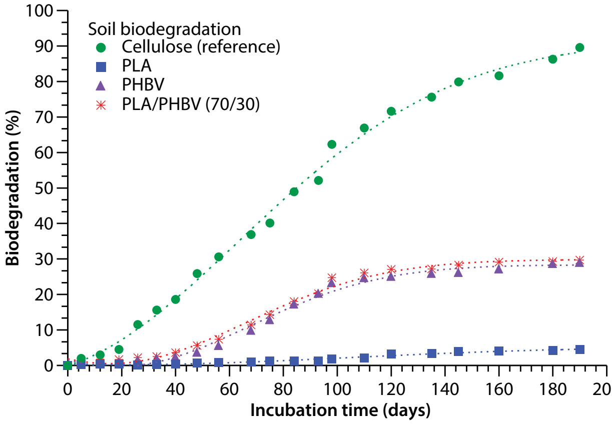


Figure 8 Biodegradation of PLA, PHBV and PLA/PHBV blend in soil burial test.

of these samples under soil burial environment. Neat PHBV and PLA/PHBV blend exhibited 35% and 32% biodegradability, respectively, after 200 days, while the hydro-biodegradable PLA material did not show appreciable microbial attack in 200 days of incubation in soil (Figure 3). Even though the PLA grade used in this study contains starch as additive, microorganisms present in the soil environment were unable to depolymerize the PLA under soil burial conditions for the incubation period of 200 days. The aerobic biodegradability of PLA has been studied by other researchers in different environments such as soil, sewage sludge and marine water [37, 41]. The results of all these studies suggest that a higher rate of biodegradability in the case of PLA can be achieved under industrial composting conditions influenced by proper pH, moisture content, temperature (58–60 °C) and thermophilic microorganisms as compared to other natural environmental conditions like soil, home compost and aqueous medium. Besides, the fact that the temperature of composting method is around the glass transition temperature of PLA (58–60 °C) which influences its hydrolytic degradation, followed by microbial (thermophilic bacteria and fungi) assimilation under composting conditions. The occurred biodegradability of PLA/PHBV (70/30) blend could be due to degradation of a polymer chain into low molecular weight compounds during the melt blending process. This study suggests that higher and faster biodegradability of biobased polymers (PLA, PHBV and PLA/PHBV blend) can be achieved under controlled composting conditions than in soil burial environments. In contrast, no fragmentation/disintegration was observed for the same test materials under soil burial conditions even after 200 days of incubation.

Table 2 presents the DSC analysis of PLA, PHBV and PLA/PHBV blends after soil burial biodegradation. The samples of PHBV and PLA/PHBV blend exhibited significant changes in their respective thermograms (melting temperature and percent crystallinity) as compared to that of PLA after 30 and 200 days incubation in soil burial (Table 2). Significant decreases in melting temperature (Tm_1 and Tm_2) and percentage crystallinity (X_c) were observed for PHBV in contrast to PLA under soil burial conditions. For the PLA/PHBV blend, a complete disappearance of a melting peak corresponding to PHBV was observed after 200 days incubation but no significant changes in melting temperature and percentage crystallinity were observed in the case of PLA (Table 2). These results indicated that the PLA, PHBV and PLA/PHBV blend had slower biodegradation rates in comparison to that in simulated composting environment.

Following incubation for a long period of 200 days, the TGA results showed a slight increase in onset temperature for all samples in comparison to that after incubation for 30 days (Table 2). These results can be attributed to the assimilation of amorphous regions of low molecular compounds in the polymer matrix by microorganisms in the first 30 days. Thereafter, the materials become crystalline (Table 2). This increases the thermal stability of the material after incubation for 30 days, which suggests that the sample has undergone slow biodegradation. Another interesting aspect to note is that the PLA/PHBV (70/30) blend has two maximum degradation peaks at temperatures of 297.7 °C and 360.0 °C which correspond to neat PHBV and PLA, respectively (Table 2). Therefore, the blend undergoes degradation behavior similar to their original components as observed in soil and compost biodegradation tests. From TGA results it was also noticed that the weight of sample residue at 600 °C was slightly increased after biodegradation. This could be due to contamination of the sample with soil/compost particles that could not be removed.

4 CONCLUSIONS

This study provided important information with respect to microbial and enzymatic attacks and physical and chemical parameters involved in the biodegradation process of biobased polymers, PLA, PHBV and PLA/PHBV blend by soil and composting methods. The results on CO_2 emission showed that PLA, PHBV and PLA/PHBV blend have a short survival time under controlled composting method in comparison to soil burial method. The blending of PLA/PHBV helps to reduce the crystalline structure of PLA, thus making the low molecular weight compounds in the blend more susceptible to undergo hydrolytic biodegradation compared to neat PLA and PHBV. The DSC, TGA, FTIR and SEM results clearly showed the biodegradation behavior and degradation pattern of biobased polymer under both soil and compost environments. The occurrence of microbial systems and conditions in the compost method provided a suitable environment for the biodegradation of biobased polymers and their blend when compared with soil burial methods.

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