Compost Science & Utilization

ISSN: (Print) (Online) Journal homepage: https://www.tandfonline.com/loi/ucsu20

Influence of Degradation of PLA with High Degree of Crystallinity on Fungal Community Structure in Compost

Mehlika Karamanlioglu & Ümit Alkan

To cite this article: Mehlika Karamanlioglu & Ümit Alkan (2020) Influence of Degradation of PLA with High Degree of Crystallinity on Fungal Community Structure in Compost, Compost Science & Utilization, 28:3-4, 169-178, DOI: 10.1080/1065657X.2020.1864514

To link to this article: https://doi.org/10.1080/1065657X.2020.1864514

COMPOST SCIENCE & UTILIZATION
2020, VOL. 28, NOS. 3–4, 169–178
https://doi.org/10.1080/1065657X.2020.1864514

Influence of Degradation of PLA with High Degree of Crystallinity on Fungal Community Structure in Compost

Mehlika Karamanlioglu[a] and Ümit Alkan[b]

[a]Department of Biomedical Engineering, Istanbul Gelisim University, Istanbul, Turkey; [b]Department of Computer Engineering, Istanbul Gelisim University, Istanbul, Turkey

ABSTRACT

Degradation rate of poly(lactic acid) (PLA), a compostable plastic, is affected by its physical properties and environmental conditions. Since PLA with different physical properties enter composting systems, investigation of degradation of PLA with strong physical properties in compost at different temperatures and its influence on compost fungal community structure are the main concerns of this study. To determine the effect of slow PLA degradation on fungal communities, PLA granules with high degree of crystallinity, 60%, were incubated in compost at 25 °C and 50 °C for 4 months at 0, 10, 25 and 50% (w/w) concentrations; their degradation rates were compared and impact of PLA degradation on compost fungal communities was examined by terminal restriction fragment length polymorphism (TRFLP). PLA granules in compost at 25 °C showed no physical changes but at 50 °C physical disintegration occurred after 4 months. TRFLP revealed that fungal community profiles in compost were affected by PLA, particularly at 50 °C where PLA degraded. Compost fungal communities in the presence of PLA at 50 °C had more variation, 63%, than at 25 °C (52%). Incubation time affected fungal community structure as during 2nd month, community structure changed specifically at 50 °C and at 50% (w/w) PLA, however, became similar to that in the absence of PLA at the end of 4th month at both temperatures indicating PLA with a high degree of crystallinity causes a temporal perturbation in compost fungal communities. In compost containing PLA at 50 °C, abundance of certain TRFs representing fungal populations increased to 30% which may involve in PLA utilization.

Introduction

Plastics have been a part of our every-day life ever since their production began in the 1950s (Barnes et al. 2009; Nithin and Goel 2017). In total, 8.3 billion metric tons of plastic have been produced since 1950s (Statista 2017a,b). By 2016, 60 million metric tons of plastic were produced in Europe and 335 million metric tons were produced globally with China being the largest plastic producers in the world (Statista 2017b). However, recycling technology has not been developed sufficiently to keep up with ever increasing amounts of plastic consumption and the array of different polymer chemistries and additives, such as plasticizers, pigments, adhesives and metals are considerable barriers for closed loop recycling (Tharanathan 2003; Vink et al. 2003; Domenek et al. 2004; Siracusa et al. 2008; Hopewell, Dvorak, and Kosior 2009). It is reported that only 9% of 8.3 billion metric tons of plastic produced globally had been recycled since 1950s and it is estimated that by 2050, 12 billion metric tons of plastic waste will be deposited in the environment (Geyer, Jambeck, and Law 2017; Statista 2017a,b). Recycling is also a costly process (Claro et al. 2016; Wyser, Leterrier, and Månson 2000). Therefore, finding alternatives to recycling of plastics such as using compostable plastics has been gaining attention in the recent years. Compostable plastics are decomposed to nontoxic residues by biological activities and PLA is a compostable and a biodegradable plastic with a potential to replace non-biodegradable plastics when used as disposable containers. Moreover, PLA has high mechanical strength comparable to non-degradable plastics (Rafael Auras, Harte, and Selke 2004; Lunt 1998; Claro

CONTACT Mehlika Karamanlioglu mkaramanlioglu@gelisim.edu.tr Faculty of Engineering and Architecture, Department of Biomedical Engineering, Istanbul Gelisim University, Cihangir Mah. Dolum Tesisleri, Murat Sengoz Sokak No. 1 Avcılar, Istanbul, Turkey.

et al. 2016). Biocomposites of PLA have been produced to improve mechanical, thermal and physical properties of PLA (González et al. 2019). Mechanical properties of PLA are influenced by its optical composition, the presence and ratio of D and L-isomers and degree of crystallinity which lead to have an effect on PLA degradation rate as when D-isomer is introduced to its structure, the optical purity of PLA decreases which also decreases crystallinity (Saha and Tsuji 2006; Höglund, Odelius, and Albertsson 2012). Lower degree of crystallinity leads to PLA degradation rate increase (Kale, Auras, and Singh 2007; Henton et al. 2005; Urayama, Kanamori, and Kimura 2002). Crystallinity gives more ordered structure to a polymer, therefore, water diffusion and hydrolysis of polymers decrease. Hence, increasing crystallinity of PLA increases strength of PLA which slows degradation rate of PLA (Vert, Mauduit, and Li 1994). Environmental factors such as temperature are also an important environmental factor to determine PLA degradation rate. Since PLA is compostable, it is degraded by compost microorganisms under humid conditions and temperatures higher than 40 °C (Karamanlioglu and Robson 2013; Sangwan and Wu 2008; Ghorpade, Gennadios, and Hanna 2001). It has previously been shown that thermophilic fungi decomposes PLA during composting (Karamanlioglu, Houlden, and Robson 2014). However, the impact of degradation of PLA with different physical properties on compost microbial communities has not been studied extensively. Since PLA can be used as an alternative to non-biodegradable plastics, understanding its effect on fungal compost communities during composting is an important parameter. In our previous study, we examined the impact of PLA degradation on compost fungal communities using a PLA source with a 35% of degree of crystallinity which was completely degraded at 50 °C after 2 months (Karamanlioglu, Preziosi, and Robson 2017). Fungal community structure returned to community structure of compost in the absence of PLA after a temporary shift when PLA films were at 10–25% (w/w) concentration. However, when PLA was 50% (w/w) in compost, fungal community structure and diversity changed due to acidification of compost (Karamanlioglu, Preziosi, and Robson 2017). In the present study, we investigated the degradation of PLA with a higher degree of crystallinity, 60%, over time at 25 °C and 50 °C; and the effect of slower degradation of PLA at different concentrations on fungal community structure in compost.

Materials and methods

Poly(lactic acid)

High molecular weight polylactic acid (PLA) granules (Mw > 150,000 g/mol) with a diameter of 5 mm were obtained from Goodfellow Cambridge Ltd, UK. According to the manufacturer, PLA granules had a glass transition temperature of ca. 55 °C, a density of 1.24 g/cm^3 and crystallinity of 60–70%.

Compost

Commercial mature compost was purchased from the Compost Shop, UK without any pretreatment and stored in sealed containers at room temperature. Compost was in its cooling phase which had a temperature of 44–46 °C. Prior to use, compost was screened to 2 mm in order to eliminate particles >2 mm. Water holding capacity (WHC) of compost was determined as 60–64% (Forster 1995) and the moisture content of compost was set to 30–32% prior to burial of PLA granules.

Preparation of compost with PLA granules

PLA granules were sterilized at 121 °C for 3 h. Sterility was checked by incubating PLA granules on PDA and LB plates. PLA granules were mixed with compost 0, 10, 25 and 50% (w/w) as previously described in (Karamanlioglu, Preziosi, and Robson 2017). Compost batches of 3 L with depth of ca. 7 cm were incubated at 25 °C and 50 °C for 4 months. During the experiment, compost boxes were weighed periodically to adjust moisture content by addition of sterile water.

pH measurement

Compost pH was measured as described previously (Forster 1995; Karamanlioglu, Preziosi, and Robson 2017). Briefly, compost sample and distilled water

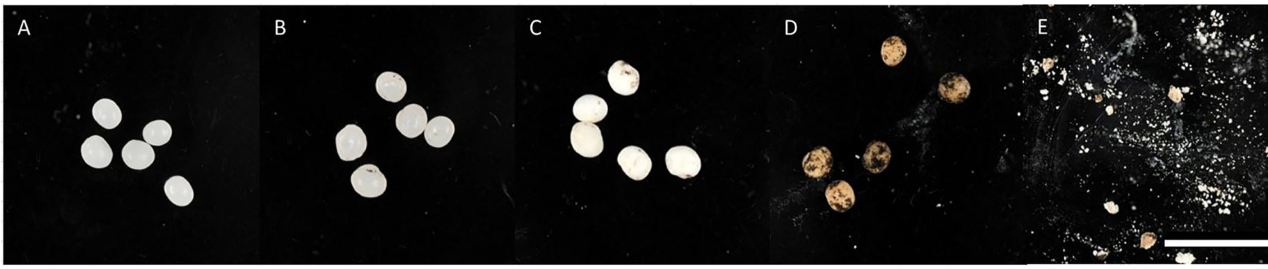


Figure 1. Morphology of PLA granules. (A) Before incubation in compost, (B) after 4 months' incubation in compost at 25 °C, (C) after 2 months' incubation in compost at 50 °C, (D) after 4 months' incubation in compost at 50 °C, (E) after 4 months' incubation with compost at 50 °C and granules after 4 months' incubation with compost at 50 °C after any physical impact. Scale bar represents 2 cm.

was mixed in 1:2 ratio and left standing for 1 h and pH was measured with a digital pH meter (Metler Toledo, UK). Compost pH was measured initially and during the 2nd and 4th month of incubation in all compost samples.

Total DNA extraction from compost

Total DNA from compost was extracted as described in our previous study (Karamanlioglu, Preziosi, and Robson 2017). Compost samples of three replicates were taken from compost batches containing 0, 10, 25, 50% (w/w) PLA granules, however, PLA granules were excluded. Power Soil DNA extraction kit (MoBio Laboratories, UK) was used to extract DNA from compost and Nanodrop 1000 (Thermo Fisher Scientific, UK) was used to quantify DNA.

Terminal restriction fragment length polymorphism (TRFLP) analysis

Influence of PLA, temperature and incubation time on fungal compost community structure was assessed by TRFLP analysis. Compost samples were removed from compost batches containing 0, 10, 25 and 50% (w/w) PLA granules before incubation and after 2 and 4 months of incubation at 25 and 50 °C. TRFLP profiles were determined as previously described (Karamanlioglu, Houlden, and Robson 2014; Karamanlioglu, Preziosi, and Robson 2017; Kawasaki, Watson, and Kertesz 2012). In brief, initially PCR amplification of ITS1-5.8S-ITS2 region of fungal rDNA was achieved by using fluorescently labeled primers ITS4-HEX (hex-TCCTCCGCTTATTGATATGC) and ITS5-FAM (fam-GGAAGTAAAAGTCGTAACAAGG) (White, Bruns, and Lee 1990) and purified amplicons (300 ng μl^{-1}) were digested using HhaI (Fermentas, 10 U/μl). Digested products were mixed with Hi-Di Formamide (Applied Biosystems, Foster City, USA) to denature DNA. GeneScan^{TM}-500 LIZ® (Applied Biosystems, Foster City, USA) was used as a size standard. Capillary electrophoresis (ABI Prism® 3100 Genetic Analyzer, Applied Biosystems Inc., Foster City, USA) was used to separate and detect fluorescently labeled fragments provided by The University of Manchester Sequencing Facility. TRF sizes were determined by comparison with the size standard using Peak Scanner^{TM} software (Applied Biosystems, Inc., California, USA). TRFs (terminal restriction fragment) representing different fungal populations were aligned by T-Align program (T-Align 2012.). T-align generated a single consensus profile for the replicates of one compost sample and then compared consensus profiles of different samples generating a TRF list with their relative fluorescence intensity (Smith et al. 2005). TRF profiles were compared by Principal Component Analysis (PCA). Diversity index (H') and evenness values (J') were determined by MVSP software (Ver. 3.12d, Kovach Computing Services) (Atlas and Bartha 1986).

Results

Physical changes of PLA granules under composting conditions over 4 months of incubation

PLA granules incubated in compost at 25 °C and 50 °C were visually evaluated. PLA granules incubated in compost at 25 °C over 4 months showed no discernible physical changes and appeared identical to control granules (Figure 1A and 1B).

Table 1. Influence of PLA granule concentration (0, 10, 25, 50%; w/w) on pH of compost after 2 and 4 months of incubation at 25 °C and 50 °C.

	Conditions/temperature		PLA granule concentration in compost 0%	10%	25%	50%
pH of compost	Initial		7.1 ± 0.2	7.1 ± 0.2	7.1 ± 0.2	7.1 ± 0.2
	25 °C	2 months	7.4 ± 0.01	7.4 ± 0.01	7.4 ± 0	7.3 ± 0.03
		4 months	6.4 ± 0.01	6.4 ± 0	6.9 ± 0.01	6.9 ± 0.02
	50 °C	2 months	7.1 ± 0.1	6.8 ± 0.02	6.9 ± 0.04	7.1 ± 0.02
		4 months	6.6 ± 0.02	6.7 ± 0.04	6.8 ± 0.05	7.1 ± 0.02

Note. Means represent three replicates ± SEM.

When incubated in compost at 50 °C, granules remained intact after 2 months but the surface appeared 'rough' (Figure 1C). After 4 months' incubation, color of granules changed (Figure 1D) and was very brittle as upon any physical contact, granules disintegrated (Figure 1E).

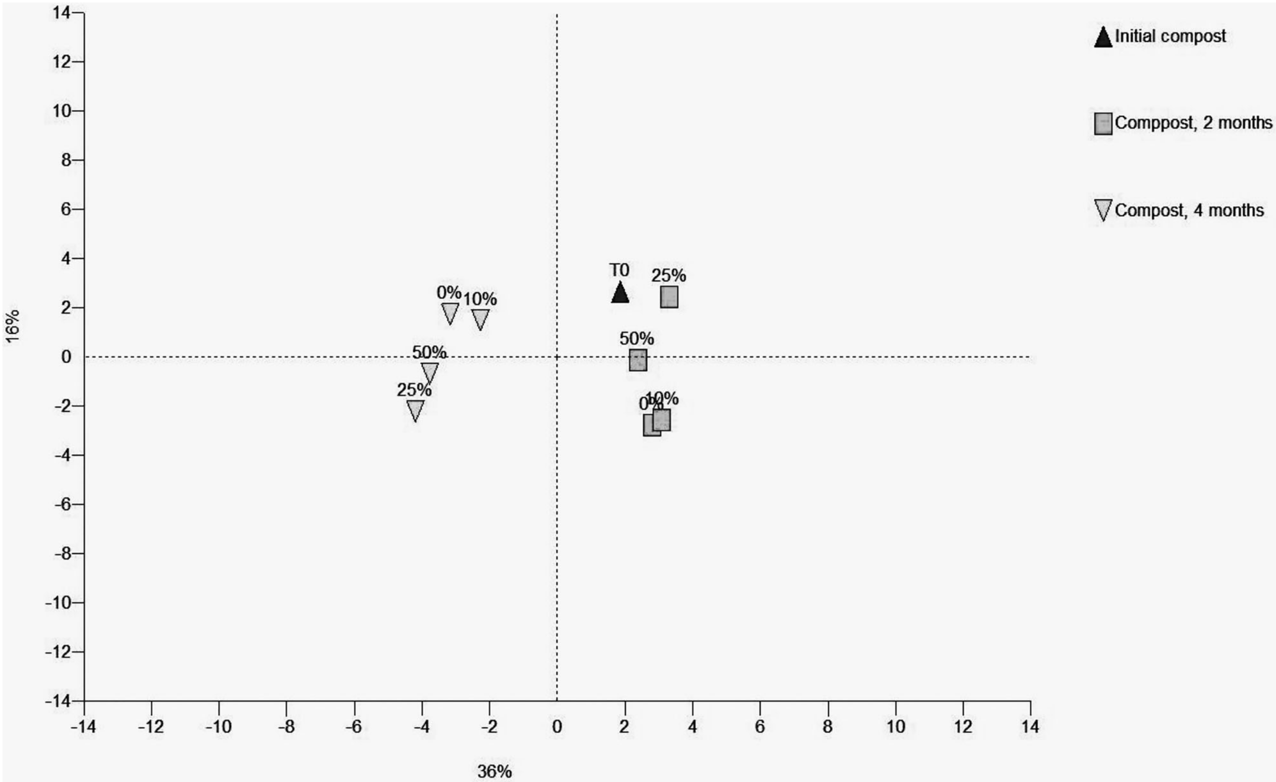
Determination of compost pH under different conditions over time

Acidity of compost containing 0, 10, 25 and 50% (w/w) PLA granules incubated at 25 °C and 50 °C were measured before and after 2 and 4 months of incubation in compost since lactic acid, the monomer of PLA, is released as a result of PLA degradation (Table 1). Initial compost pH was 7.1 ± 0.2. Neither at 25 °C nor at 50 °C, no significant ($P > 0.05$) change in pH was observed in compost in the presence of PLA granules after 4 months of incubation (Table 1).

Determination of fungal community structure change upon PLA granules' degradation under different conditions over time

TRF profiles from compost incubated at 25 and 50 °C for 2 and 4 months containing 0, 10, 25 and 50% (w/w) PLA granules were analyzed and compared with initial fungal compost community by principle component analysis (PCA) after normalization (Figure 2). Figure 2A shows that when PLA granules were incubated at 25 °C and principal components, PC1 and PC2, showed 52% variability in total within the dataset. PC1 accounted for 36% of the variation and showed differences between the group comprising initial compost and compost containing 0–50% PLA granules after 2 months of incubation; and the group comprising compost containing 0–50% PLA granules incubated for 4 months. PC2 accounted for 16% of the variation mainly showing the difference between the group of initial compost with compost containing 25% (w/w) PLA granules during the 2nd month; and the group of compost containing 10% (w/w) and no PLA granules during the 2nd month. Although fungal community profiles at 25 °C changed over time at each PLA concentration level, the community change was due to incubation time rather than PLA concentration as all compost samples containing PLA clustered closely with compost containing no PLA at each time period. When PLA granules were incubated in compost at 50 °C, PC1 and PC2 showed 63% of the variability within the dataset (Figure 2B). PC1 accounted for 40% of the total variability and highlighted the difference between compost containing 50% PLA granules incubated for 2 months from the rest of the compost samples, while PC2 explained 23% of the total variability and mainly showed the difference between compost containing 10% PLA granules incubated for 2 months from compost with 50% granules incubated for 2 months. The largest effect on the community profiles was observed in the presence of PLA granules after 2 months' incubation as they cluster away from the initial compost and following 2 months' incubation in the absence of PLA as well as separating from each other. However, after 4 months of incubation, community profiles in the presence of PLA grouped together close to the compost community incubated for 4 months in the absence on PLA.

The influence of PLA granule concentrations (0, 10, 25, 50%, w/w) on compost fungal diversity was evaluated by calculating the number of TRFs, Shannon index (H') and evenness (J') corresponding to richness, diversity and sizes of populations, respectively (Table 2). When PLA granules were incubated at 25 °C for 2 months, no significant trend with respect to PLA



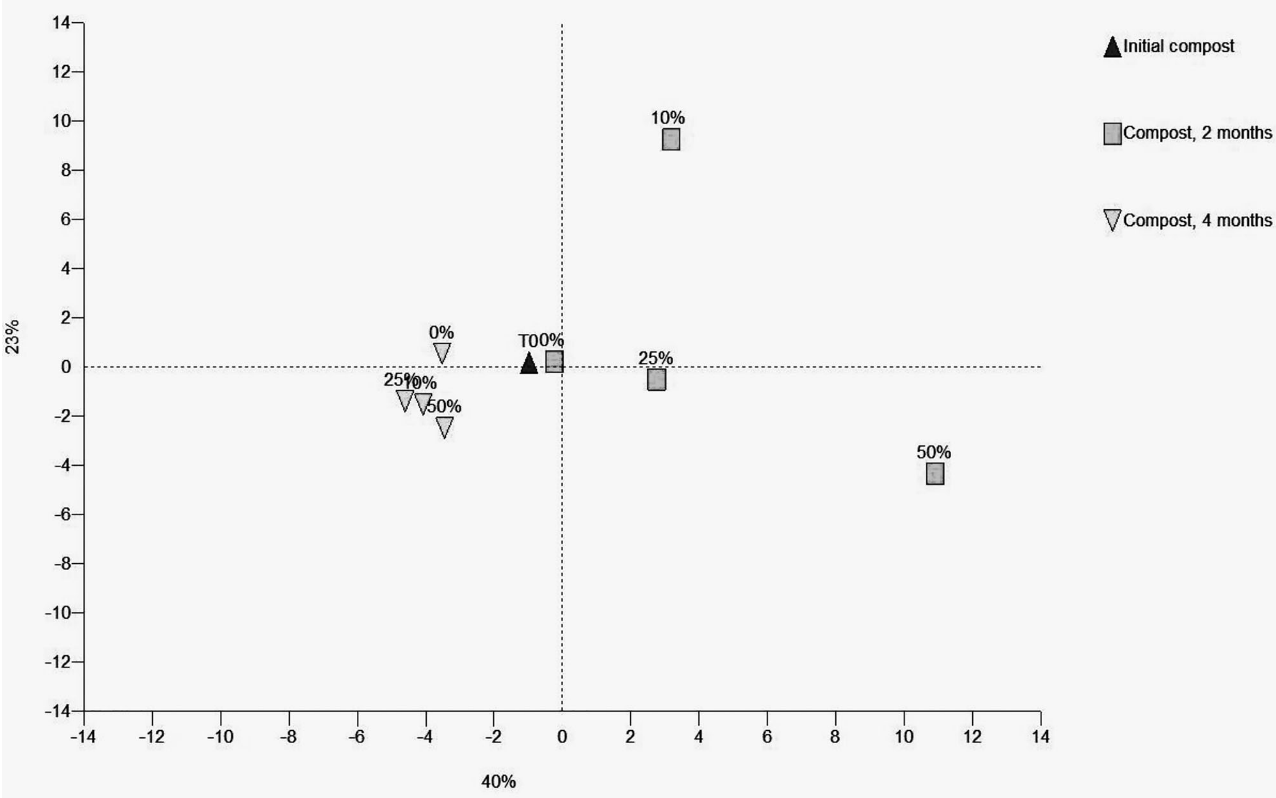


Figure 2. Principal Component Analysis (PCA) of TRFLP profiles of fungal communities isolated from initial compost and compost containing 0–50% (w/w) PLA granules (A) at 25 °C and (B) at 50 °C after incubation for 2 and 4 months.

Table 2. Diversity index (H'), evenness (J'), number of TRFs and % of PLA unique TRFs of fungal communities in compost containing 0–50% (w/w) PLA granules.

		PLA% in compost	Shannon diversity (H')	Evenness (J')	Number of TRFs	% of PLA unique TRFs
		0%, Initial compost	3.6	0.7	145	
25 °C, compost	2 months	**0%**	2.9	0.6	129	
		10%	2.5	0.5	162	6
		25%	1.9	0.5	60	2
		50%	2.7	0.5	140	1
	4 months	**0%**	3.4	0.7	149	
		10%	3.3	0.7	140	7
		25%	3.6	0.7	162	4
		50%	3.6	0.7	175	10
50 °C, compost	2 months	**0%**	3.2	0.7	85	
		10%	2.7	0.6	107	22
		25%	3.0	0.6	162	28
		50%	2.3	0.5	106	28
	4 months	**0%**	3.7	0.8	138	
		10%	3.4	0.7	148	26
		25%	3.4	0.7	144	23
		50%	3.4	0.6	207	38

Note. Three replicates which were pooled after PCR were used for each compost sample. Two technical replicates were aligned to represent each compost sample.

concentration was observed on the compost fungal community in terms of number of TRFs, diversity and evenness of the compost samples. On the 4th month of incubation, the number of TRFs, diversity and evenness increased compared to 2nd month regardless of the PLA concentrations. As PLA granules were incubated at 50 °C, although the TRF numbers were more in compost with 10, 25 and 50% (106–162) than 0% PLA containing compost (85); no discernible PLA concentration effect was observed after 2 months of incubation. The number TRFs, diversity and evenness were more on the 4th month than 2nd month. At 10 and 25% (w/w) of PLA granule concentration, number of TRFs (≃148), H' (3.4) and J' (0.7) were similar with compost in the absence of PLA (TRFs = 138; H' = 3.7; J' = 0.8) after 4 months of incubation at 50 °C. At 50% (w/w) of PLA granule concentration, evenness (0.6) was less and the number of TRFs (207) were more than those of other PLA concentrations (138–148 TRFs) indicating increase of certain fungal populations in compost.

The number and distribution of TRFs in compost with PLA granules at 25 and 50 °C were represented as peaks with their length in base pairs (bp) and relative abundance (peak height) in Figure 3. The number of TRFs that were present only in compost containing PLA (unique TRFs) was also determined (Table 2). These TRFs were present in compost in the presence of PLA but not detectable either in the initial compost community or in compost in the absence of PLA. When PLA granules were incubated at 25 °C, between 1 and 17% of TRFs were only recovered from PLA containing compost while at 50 °C, they represented between 3 and 38% of all detectable TRF's (Table 2). At 50% (w/w) of PLA granule concentration in compost, number of unique TRFs (38%) were more than the other PLA concentrations (≃25%) in compost. However, the unique TRFs all had low relative abundances (<1%). Some peaks were common in all compost communities at either 25 °C or 50 °C with relatively high abundances. For example, at 25 °C, TRFs of 89.71 bp, 119.83 bp, and 169.81 bp appeared in all communities with relative abundances ranging from 2–38% (Figure 3A). At 50 °C, the abundance of some TRFs, such as TRFs of 89.6 bp and 159.45 bp, increased in the presence of PLA in compost (Figure 3B). A TRF of 165.42 bp length had a relative abundance of only 0.02–0.1% in compost without PLA, however, its abundance increased greatly to 6–30% after 4 months of incubation in compost containing PLA granules.

Discussion

We investigated the effect of degradation of PLA with high degree of crystallinity (60%) on the fungal community in compost at different temperatures at different incubation intervals using TRFLP. Different concentrations of PLA granules (0–50% w/w) were incubated for 2 and 4 months

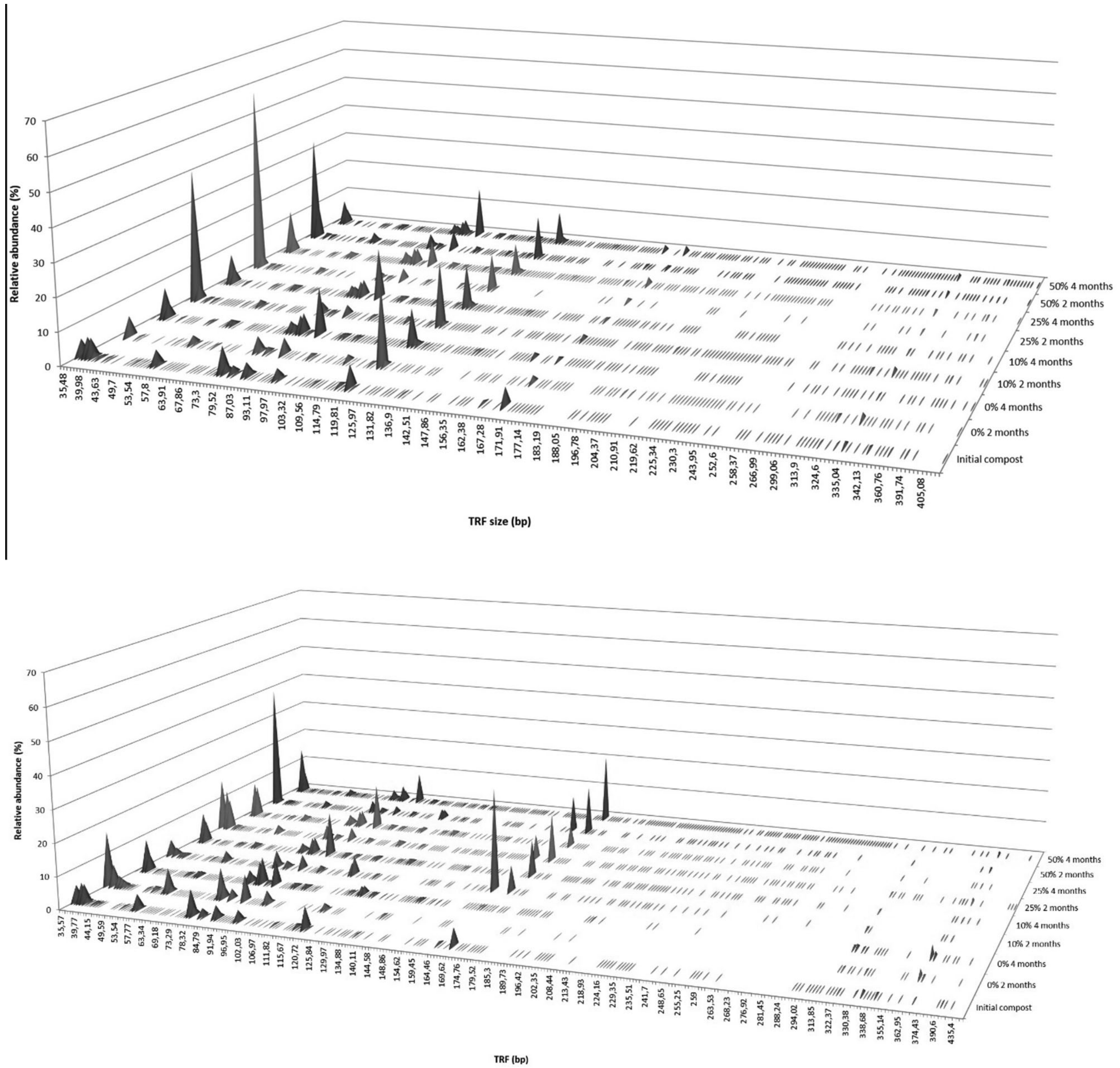


Figure 3. TRF distribution of fungal communities isolated from initial compost and compost with 0–50% PLA granules (A) at 25 °C and (B) at 50 °C on the 2nd and 4th month of incubation. Relative abundances (%) of TRFs were shown with fragment sizes, base pairs (bp).

at 25 and 50 °C. During composting, temperature of the compost varies, therefore there are different composting phases (Hellmann et al. 1997). In this research, compost 25 °C and 50 °C were selected to represent mature phase after cooling and initial cooling phase when the number of fungi increases in the compost, respectively. No degradation of PLA granules was observed at 25 °C over 4 months, however, physical disintegration of PLA granules was observed when incubated in compost at 50 °C. As reported previously, PLA degrades efficiently at temperatures close to its glass transition temperature (Tg, 55–62 °C) in humid environments (Itävaara, Karjomaa, and Selin 2002; Weir, Buchanan, and Orr 2004; Kale, Auras, and Singh 2007; Saadi et al. 2012; Agarwal, Koelling, and Chalmers 1998; Vargas et al. 2009; Karamanlioglu and Robson 2013)

PLA granules with high degree of crystallinity degraded relatively slowly as the degradation was observed at the end of 4th month of incubation when compared with the PLA films with lower degree of crystallinity (35%) from a previous study (Karamanlioglu, Preziosi, and Robson 2017). At 50 °C, PLA films degraded at a faster rate than PLA granules as films disintegrated after 2 months while granules disintegrated after 4 months (Karamanlioglu, Preziosi, and Robson 2017). The slower degradation rate of PLA granules most likely reflects differences of degree of crystallinity of PLA granules and films which is known to affect degradation rate (Nishida and Tokiwa 1992; R Auras, Lim, and Tsuji 2010). PLA granules had a higher degree of crystallinity (60–70%) than PLA films (ca. 30%). Water is less readily able to diffuse into more ordered crystalline regions within PLA compared to less ordered amorphous regions and higher crystallinity therefore decreases the rate of hydrolysis (Reeve et al. 1994; Fambri et al. 2002).

When PLA degrades, its monomer, lactic acid, is released which decreases the pH of its surrounding (Torres et al. 1996; Agarwal, Koelling, and Chalmers 1998; Karamanlioglu and Robson 2013). In this study, pH of compost did not change (Table 1) when incubated with PLA granules at 25 °C for 4 months which is consistent with the lack of physical degradation (Figure 1B). This result is consistent with the previous study as PLA films, though having lower degree of crystallization, did not show any sign of degradation after 4 months of incubation in compost at 25 °C (Karamanlioglu, Preziosi, and Robson 2017). Although PLA disintegration was observed on the 4th month of incubation at 50 °C (Figure 1D and 1E), no significant pH change occurred in compost containing PLA granules after 4 months of incubation (Table 1). Compost microorganisms would utilize PLA degradation products of PLA as a carbon source, therefore, pH decrease was not observed immediately after disintegration of PLA granules. In accordance with this, in another study, pH of a biotic reactor containing PLA increased at 40–60 °C, whereas pH of an abiotic reactor and sterile water decreased (Agarwal, Koelling, and Chalmers 1998). As degradation was detected only after 4 months, a longer period of incubation is needed to be studied in order to investigate the longer term effects of PLA degradation on pH change and on the fungal community. In our previous study same trend was observed when PLA films were buried in compost with the exception of compost of 50% (w/w) PLA films at 50 °C where significant pH decrease of compost was observed after 4 months of incubation since there was excess amount of lactic acid to be consumed by microorganisms after PLA disintegration on the 2nd month (Karamanlioglu, Preziosi, and Robson 2017). When 50% of PLA films were incubated in compost at 50 °C, PLA films were completely degraded on the 2nd month and pH decreased from 6.5 to 3.1 after 4 months of incubation (Karamanlioglu, Preziosi, and Robson 2017). As degradation of PLA granules occur at a lower rate compared to PLA films due to degree of crystallinity, the terminal release of lactic acid may not have been reached after 4 months since PLA disintegrated only after a physical impact (Figure 1D and 1E).

Principal component analysis (PCA) of TRFLP profiles (Figure 2) was used to determine the effect of different PLA granule concentrations on compost fungal community at 25 and 50 °C. During incubation with PLA granules, it was shown that fungal diversity and community structure of compost were affected by the presence of PLA at both 25 °C where little or no degradation occurred over 4 months, and at 50 °C where extensive degradation occurred.

In the presence of PLA granules, populations after 2 months of incubation showed the least similarity to the populations in the absence of PLA but those after 4 months of incubation were similar to the population in the absence of PLA suggesting that initial degradation caused a shift in the population but the population became more similar to that in the absence of PLA by 4 months. Similarly, in the presence of PLA films with the degree of crystallinity of 35%, the greatest divergence to the population compared to the absence of PLA was also seen after 2 months and became more similar to the population in the absence of PLA after 4 months of incubation (Karamanlioglu, Preziosi, and Robson 2017). However, the population containing 50% (w/w) PLA films remained divergent due to the significant pH decrease (Karamanlioglu, Preziosi, and Robson 2017). Some TRFs only appeared in compost containing PLA granules especially at 50 °C when PLA granules were disintegrated which is in accordance with our previous study where compost contained PLA films (Karamanlioglu, Preziosi, and Robson 2017). The abundance of unique TRFs was smaller than 1% indicating that they did not have an extensive role on PLA degradation. However, certain TRF relative abundance in compost increased after incubation with PLA specifically after PLA degradation at 50 °C (Figure 3). These TRFs may refer to fungal populations that utilize PLA as a carbon source and may have a role in PLA degradation. PLA with a high degree of crystallinity causes a temporal perturbation in compost fungal community structure as the pH of compost does not change which is consistent with the previous results (Karamanlioglu, Preziosi, and Robson 2017). It is essential to determine the physical properties of PLA to be used as disposable containers which enter composting systems after use. Higher degree of crystallinity decreases PLA degradation rate in compost which may affect the quality of commercial composting.

ORCID

Mehlika Karamanlioglu http://orcid.org/0000-0002-4814-6346

References

Agarwal, M., K. W. Koelling, and J. J. Chalmers. 1998. Characterization of the degradation of polylactic acid polymer in a solid substrate environment. *Biotechnology Progress* 14 (3):517–26. doi: 10.1021/bp980015p.

Atlas, R. M., and R. Bartha. 1986. *Microbial ecology: Fundamentals and applications*. Menlo Park, CA: Benjamin-Cummings Pub. Co.

Auras, R., B. Harte, and S. Selke. 2004. An overview of polylactides as packaging materials. *Macromolecular Bioscience* 4 (9):835–64. doi: 10.1002/mabi.200400043.

Auras, R., L.-T. Lim, and H. Tsuji. 2010. *Poly(lactic acid): Synthesis, structures, properties, processing, and applications*. Vol. 6. New Jersey: John Wiley & Sons, Inc.

Barnes, D. K. A., F. Galgani, R. C. Thompson, and M. Barlaz. 2009. Accumulation and fragmentation of plastic debris in global environments. *Philosophical Transactions of the Royal Society of London. Series B, Biological Sciences* 364 (1526):1985–98. doi: 10.1098/rstb.2008.0205.

Claro, P. I. C., A. R. S. Neto, A. C. C. Bibbo, L. H. C. Mattoso, M. S. R. Bastos, and J. M. Marconcini. 2016. Biodegradable blends with potential use in packaging: A comparison of PLA/chitosan and PLA/cellulose acetate films. *Journal of Polymers and the Environment* 24 (4):363–71. doi: 10.1007/s10924-016-0785-4.

Domenek, S., P. Feuilloley, J. Gratraud, M.-H. Morel, and S. Guilbert. 2004. Biodegradability of wheat gluten based bioplastics. *Chemosphere* 54 (4):551–9. doi: 10.1016/S0045-6535(03)00760-4.

Fambri, L., C. Migliaresi, K. Kesenci, and E. Piskin. 2002. Biodegradable polymers. In *Integrated biomaterials science*, ed. Rolando Barbucci, 119–87. New York: Kluwer Academic/Plenum Publishers.

Forster, J. 1995. Soil sampling, handling, storage and analysis. In *Methods in applied soil microbiology and biochemistry*, ed. Kassem Alef and Paolo Nanniperi, 49–121. London: Academic Press.

Geyer, R., J. R. Jambeck, and K. L. Law. 2017. Production, uses, and fate of all plastics ever made. *Science Advances* 3 (7):e1700782. doi: 10.1126/sciadv.1700782.

Ghorpade, V. M., A. Gennadios, and M. A. Hanna. 2001. Laboratory composting of extruded poly(lactic acid) sheets. *Bioresource Technology* 76 (1):57–61. doi: 10.1016/S0960-8524(00)00077-8.

González, M. E., L. Aida, A. Pérez Fonseca, E. O. Cisneros, L. Ricardo, M. González, D. E. Ramírez, A. Denis, R. Jorge, and R. Robledo Ortíz. 2019. Effect of maleated PLA on the properties of rotomolded PLA-agave fiber biocomposites. *Journal of Polymers and the Environment* 27 (1):61–73. doi: 10.1007/s10924-018-1308-2.

Hellmann, B., L. Zelles, A. Palojarvi, and Q. Bai. 1997. Emission of climate-relevant trace gases and succession of microbial communities during open-windrow composting. *Applied and Environmental Microbiology* 63 (3):1011–8. doi: 10.1128/AEM.63.3.1011-1018.1997.

Henton, D. E., P. Gruber, J. Lunt, and J. Randall. 2005. Polylactic acid technology. *Natural Fibers, Biopolymers, and Biocomposites* 16:527–78.

Höglund, A., K. Odelius, and A.-C. Albertsson. 2012. Crucial differences in the hydrolytic degradation between industrial polylactide and laboratory-scale poly(L-lactide). *ACS Applied Materials & Interfaces* 4 (5):2788–93. doi: 10.1021/am300438k.

Hopewell, J., R. Dvorak, and E. Kosior. 2009. Plastics recycling: Challenges and opportunities. *Philosophical Transactions of the Royal Society of London. Series B, Biological Sciences* 364 (1526):2115–26. doi: 10.1098/rstb.2008.0311.

Itävaara, M., S. Karjomaa, and J.-F. Selin. 2002. Biodegradation of polylactide in aerobic and anaerobic thermophilic conditions. *Chemosphere* 46 (6):879–85. http://www.ncbi.nlm.nih.gov/pubmed/11922068. doi: 10.1016/S0045-6535(01)00163-1.

Kale, G., R. Auras, and S. P. Singh. 2007. Comparison of the degradability of poly (lactide) packages in composting and ambient exposure conditions. *Packaging Technology and Science* 20 (1):49–70. doi: 10.1002/pts.742.

Karamanlioglu, M., A. Houlden, and G. D. Robson. 2014. Isolation and characterisation of fungal communities associated with degradation and growth on the surface of poly(lactic) acid (PLA) in soil and compost. *International*

Biodeterioration & Biodegradation 95:301–10. doi: 10.1016/j.ibiod.2014.09.006.

Karamanlioglu, P., R. Preziosi, and G. D. Robson. 2017. The compostable plastic poly(lactic) acid causes a temporal shift in fungal communities in maturing compost. *Compost Science and Utilization* 25 (4):211–19. doi: 10.1080/1065657X.2016.1277808..

Karamanlioglu, M., and G. D. Robson. 2013. The influence of biotic and abiotic factors on the rate of degradation of poly (lactic) acid (PLA) coupons buried in compost and soil. *Polymer Degradation and Stability* 98 (10):2063–71. doi: 10.1016/j.polymdegradstab.2013.07.004.

Kawasaki, A., E. R. Watson, and M. A. Kertesz. 2012. Indirect effects of polycyclic aromatic hydrocarbon contamination on microbial communities in legume and grass rhizospheres. *Plant and Soil* 358 (1–2):169–82. doi: 10.1007/s11104-011-1089-z.

Lunt, J. 1998. Large-scale production, properties and commercial applications of polylactic acid polymers. *Polymer Degradation and Stability* 59 (1–3):145–52. doi: 10.1016/S0141-3910(97)00148-1.

Nishida, H., and Y. Tokiwa. 1992. Effects of higher-order structure of poly (3-hydroxybutyrate) on its biodegradation. I. Effects of heat treatment on microbial degradation. *Journal of Applied Polymer Science* 46 (8):1467–76. Springer: doi: 10.1002/app.1992.070460818.

Nithin, B., and S. Goel. 2017. Degradation of plastics. In *Advances in solid and hazardous waste management*, 235–47. Cham, Switzerland: Springer International Publishing.

Reeve, M. S., S. P. McCarthy, M. J. Downey, and R. A. Gross. 1994. Polylactide stereochemistry: Effect on enzymic degradability. *Macromolecules* 27 (3):825–31. doi: 10.1021/ma00081a030.

Saadi, Z., A. Rasmont, G. Cesar, H. Bewa, and L. Benguigui. 2012. Fungal degradation of poly(L-lactide) in soil and in compost. *Journal of Polymers and the Environment* 20 (2):273–82. doi: 10.1007/s10924-011-0399-9.

Saha, S. K., and H. Tsuji. 2006. Effects of molecular weight and small amounts of D-lactide units on hydrolytic degradation of poly (L-lactic acid)s. *Polymer Degradation and Stability* 91 (8):1665–73. doi: 10.1016/j.polymdegradstab.2005.12.009.

Sangwan, P., and D.-Y. Wu. 2008. New insights into polylactide biodegradation from molecular ecological techniques. *Macromolecular Bioscience* 8 (4):304–15. doi: 10.1002/mabi.200700317.

Siracusa, V., P. Rocculi, S. Romani, and M. D. Rosa. 2008. Biodegradable polymers for food packaging: A review. *Trends in Food Science & Technology* 19 (12):634–43. doi: 10.1016/j.tifs.2008.07.003.

Smith, C. J., B. S. Danilowicz, A. K. Clear, F. J. Costello, B. Wilson, and W. G. Meijer. 2005. T-align, a web-based tool for comparison of multiple terminal restriction fragment length polymorphism profiles. *FEMS Microbiology Ecology* 54 (3):375–80. http://www.ncbi.nlm.nih.gov/pubmed/16332335. doi: 10.1016/j.femsec.2005.05.002.

Statista. 2017a. *Global plastic production*. Accessed July 24, 2018. https://www.statista.com/statistics/282732/global-production-of-plastics-since-1950/.

Statista. 2017b. *Selected data on plastic production and waste worldwide 2017*. Accessed July 24, 2018. https://www.statista.com/statistics/728466/plastic-production-and-waste-worldwide-2017/.

T-Align. 2012. *T-align, a web based tool for comparison of multiple terminal restriction fragment length polymorphism profile*. Accessed January 1, 2013. http://inismor.ucd.ie/~talign/.

Tharanathan, R. N. 2003. Biodegradable films and composite coatings: Past, present and future. *Trends in Food Science & Technology* 14 (3):71–8. doi: 10.1016/S0924-2244(02)00280-7.

Torres, A., S. M. Li, S. Roussos, and M. Vert. 1996. Poly(lactic acid) degradation in soil or under controlled conditions. *Journal of Applied Polymer Science* 62 (13):2295–302. doi: 10.1002/(SICI)1097-4628(19961226)62:13 < 2295::AID-APP14 > 3.0.CO;2-2.

Urayama, H., T. Kanamori, and Y. Kimura. 2002. Properties and biodegradability of polymer blends of poly (l-lactide) s with different optical purity of the lactate units. *Macromolecular Materials and Engineering* 287 (2):116–21. doi: 10.1002/1439-2054(20020201)287:2<116::AID-MAME116>3.0.CO;2-Z.

Vargas, L. F., B. A. Welt, P. Pullammanappallil, A. A. Teixeira, M. O. Balaban, and C. L. Beatty. 2009. Effect of electron beam treatments on degradation kinetics of polylactic acid (PLA) plastic waste under backyard composting conditions. *Packaging Technology and Science* 22 (2):97–106. doi: 10.1002/pts.832.

Vert, M., J. Mauduit, and S. Li. 1994. Biodegradation of PLA/GA polymers: Increasing complexity. *Biomaterials* 15 (15):1209–13. doi: 10.1016/0142-9612(94)90271-2.

Vink, E. T. H., K. R. Rábago, D. A. Glassner, and P. R. Gruber. 2003. Applications of life cycle assessment to NatureWorks™ polylactide (PLA) production. *Polymer Degradation and Stability*. 80 (3):403–19. doi: 10.1016/S0141-3910(02)00372-5.

Weir, N. A., F. J. Buchanan, and J. F. Orr, 2004. Degradation of poly-L-lactide. Part 1: In vitro and in vivo physiological temperature degradation. *Proceedings of the Institution of Mechanical Engineers. Part H, Journal of Engineering in Medicine* 218 (5):307–19. http://www.ncbi.nlm.nih.gov/pubmed/15532996. doi: 10.1243/0954411041932782.

White, T. J., T. Bruns, and S. Lee. 1990. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In *PCR protocols: A guide to methods and applications*, ed. M. A. Innis, D. H. Gelfand, J. J. Sninsky, and T. J. White, 315–22. San Diego, CA: Academic Press.

Wyser, Y., Y. Leterrier, and J.-A. E. Månson. 2000. Effect of inclusions and blending on the mechanical performance of recycled multilayer PP/PET/SiOx films. *Journal of Applied Polymer Science* 78 (4):910–8. doi: 10.1002/1097-4628(20001024)78:4<910::AID-APP260>3.0.CO;2-O.