

Microbial Community Changes During the Composting of Municipal Solid Waste

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ABSTRACT

Phospholipid fatty acid (PLFA) analysis has been used to characterize microorganisms from a range of different environments, but has not been previously used in the assessment of compost organisms. Compost processing and maturity are assumed to be related to the microorganisms present, but methods to elucidate and evaluate these relationships are lacking. In this study, PLFA analysis was used to follow microbial community changes during the composting of municipal solid waste (MSW). Patterns of change were compared between pilot- and full-scale facilities and between varied feedstocks. At the pilot level, actual MSW and two synthetic MSW formulations (similar C:N, different available C) were composted. At the full-scale facilities, actual MSW was composted as was actual MSW amended with nitrogen. The PLFA data generated by all studies was analyzed using principal component and multivariate statistical methods. The PLFA profiles changed over the composting process in a consistent and predictable manner. PLFA profiles also proved to be characteristic of specific stages of composting and may, therefore, be useful in evaluating (and optimizing) the progress of material processing and product maturity.

Introduction

As composting becomes an accepted form of waste management in the United States, a better understanding of microbial community dynamics is needed in order to improve and evaluate material processing and quality. Many early studies dealt with the isolation and description of various compost microorganisms [7, 8, 24, 26]. Much of this work was based on the assumption that an organism could be found and

utilized to speed up decomposition [6, 7]. This hunt for a magic inoculum now appears futile, as it is unlikely that any particular organism could be the chief agent of decomposition throughout the variable conditions that occur over the composting process [8, 18].

Successful composting appears to be a result of the ability of the system to supply the microbial community with its basic needs: moisture, oxygen, temperature control, and adequate mixing. With these conditions satisfied, the microbial community follows a predictable successional pattern. The first group is dominated by the mesophilic zymogenous organisms; the next one by thermophilic organisms. The final

community is comprised, primarily, of mesophilic autochthonous organisms. These general communities are made up of specific microorganisms. The initial decomposers of organic wastes are the fungal and bacterial components. During the subsequent thermophilic phase, the actinomycetes appear and the fungal populations decline. In the final phase of composting, curing, the actinomycetes remain and the fungi reappear along with cellulose-decomposing bacteria [11, 12, 13, 21].

This study employed phospholipid fatty acid analysis (PLFA) to follow the microbial community during the composting of MSW, nitrogen-amended MSW, and synthetic MSW materials. Composting was accomplished at both full- and pilot-scale facilities. To eliminate the selectional bias associated with plating and other microbial isolation methods, a direct lipid extraction was used to characterize the microbial community shifts [9, 10, 27, 28]. Although not previously used for compost, PLFA analysis has been used to characterize the microbial communities of such diverse systems as those in marine sediments, soils, and plant rhizospheres [4, 22, 23, 27].

Materials and Methods

Feedstock

Actual MSW was obtained from a full-scale composting facility (Recomp, St. Cloud, Minn). It consisted of mixed residential waste devoid of yard wastes. Preprocessing of the MSW included screening the material to seven inches and then drum homogenizing it for three days. Material was again decreased (to 1.5 inches) and then either diverted to a pilot-scale facility (Procter & Gamble, Cincinnati, Ohio), or fed into a forced aeration trench (Royer Industries Inc., Kingston, Pa.) at the Recom full-scale facility.

Synthetic MSW consisted of two formulations of newspaper, rabbit food, composted cow manure, and corn cob pellets. These two formulations are referred to as the first and second synthetic. These materials were provided and processed by the Procter and Gamble composting lab. The primary difference between the two is an increase in the available carbon in the second formulation. The percent newspaper, rabbit food, cow manure, and corn cobs, respectively, in the first synthetic mixture was 30, 40, 30, and 0. In the second synthetic, the percentages were 3, 52, 1, and 44. These resulted in C:N ratios of 29:1 (first synthetic) and 36:1 (second synthetic). Details of these materials are also reported by Cook, et al. [3].

Pilot-Scale Facility

Composting at the pilot-scale facility was performed in 207-liter (55 gal) drums. Feedstocks used were the actual MSW and the synthetic MSWs. Aeration was done by temperature feedback

(turned on when temperatures exceeded 60°C), as well as by a computer-controlled, timed aeration. Material in the drums was turned and the moisture levels adjusted to approximately 50% every three days. Samples were taken during the turning process and transported on ice to the University of Cincinnati for analysis.

Full-Scale Facility

At the Recom facility, MSW was placed into rotary drums for mixing and then screening to 38 mm (1.5 inches). The composting process was initiated by placing this screened material into Royer trenches (Royer Industries Inc., Kingston, Pa.), that were approximately 40 m long, 3.4 m wide, and 3 m deep. Throughout the process, these trenches were aerated and cooled (to 65°C) from underneath with forced air. Material was loaded into the front of a trench, mixed, and watered every three days. Mixing was accomplished by an overhead apparatus that moved toward the front (loading) end of the trench while throwing the compost on a conveyor and dropping it 4 m behind. In this manner, it took approximately 52 to 62 days for any one load to progress to the end of the trench. The compost was then screened by ballistic separation to remove unwanted particles and placed on a curing pad in 4 m high windrows.

At the full-scale facility two different types of studies were conducted. During the first study, three trenches were sampled at five different points; these locations were chosen to represent five different time points in the MSW composting process. The second, separate study consisted of following two MSW loadings throughout the composting process. The first trench functioned as a control to the second trench, where 22.68 kg of nitrogen, in the form of ammonium nitrate, was added (at the front end) to 12.7 tons of MSW.

During both studies, samples were taken by removing 2 liters of material from three equidistant positions across a 3.4-meter trench and at a depth of 1 m. The three samples were mixed together and transported on ice to the University of Cincinnati for analysis. A 408.24-kg subsample from the 27.21-metric ton run of the control, nitrogen amended, and unamended composts, was sent to the pilot-scale facility, upon exiting the trench, for monitoring of curing.

Sample Identification

Samples are identified as designated in Table 1. In this manuscript's figures, samples are also given final numbers corresponding to the relative age of the material, "1" being early in the process and "7" being during curing. These numbers allow a relative comparison to be made (between facilities) across the composting process, as the actual number of days for completion varied between the full- (90 to 365 days) and pilot- (45 days) scale systems, but not within. In figures, the individual compost runs are identified by small letter subscripts to the material age. An example of a full sample identification code could be: P/MSW⁰/1_a. This would indicate pilot-scale, actual MSW not N amended, immature material, run a.

PLFA Analysis

Prior to analysis, compost samples were briefly hand sorted to remove large inert material, such as plastics and glass. Total lipid

Table 1. Sample identification codes for various feed materials composted at the full- or pilot-scale

Sample ID	Scale	Feed material
P/MSW	Pilot	Actual MSW
P/S	Pilot	First synthetic MSW
P/SS	Pilot	Second synthetic MSW
F/MSW	Full	Actual MSW
F/MSW ⁰	Full	Actual MSW without added N
F/MSW ⁺	Full	Actual MSW with added N

extraction was performed on 15 g of compost by a modified Bligh and Dyer method [1, 16, 25, 27]. The total lipids were extracted into the chloroform layer, dried under nitrogen, frozen (-20°C) and shipped to Microbial Insights, Inc. (Knoxville, Tenn.) for GC/MS analysis of microbial phospholipid fatty acids (PLFA) [27, 28].

The PLFA nomenclature used in this study follows the pattern of A:B ω C. The “A” position identifies the total number of carbon atoms in the fatty acid. Position “B” is the number of double bonds from the aliphatic (ω) end of the molecule. “C” designates the carbon atom from the aliphatic end before the double bond. This is followed by a “c” for *cis* or a “t” for *trans* configuration of monoenoics. The prefixes “i” and “a” stand for *iso* or *anteiso*, respectively. Midchain branching is noted by “me,” and cyclopropyl fatty acids are designated as “cy.”

Statistical analysis focused on the samples and their PLFAs. Characterization of the microbial communities present in the samples was based on the identification of the PLFAs.

Statistical Analysis

PLFA data (mole percent and total lipid phosphate) were divided into classes based on the method of PLFA formation: terminally branched saturates; monoenoics; polyenoics; midchain branched saturates; and normal saturates. The data set was transformed by removing all fatty acids that were less than 1% of the profile. The fatty acids of 16:0 and 18:0 were also removed since these PLFAs are ubiquitous in the microbial community and are found in large quantities [1, 5, 10]. Analyses of the fatty acid profiles of the compost samples were initially performed on raw ($\mu\text{mole PLFA g}^{-1}$ dry weight MSW) values and mole percentages. Since, in this preliminary analysis, either expression of PLFA data yielded similar results, subsequent analysis and discussions relied on the mole percentage data.

After the PLFA data transformations were completed, the compost samples were subjected to principal component and incremental cluster analysis using Ein*Sight 3.0 (Infometrix, Inc.). Principal component analysis simultaneously considers many correlated variables (here PLFAs) and identifies the lowest number needed to accurately represent the structure of the data set. These variables are then linearly combined with the eigenvectors of the correlation matrix to generate a principal component axis. The first principal component axis (P1) is formed from the original variables with the greatest variance. All subsequent principal components

(P2, P3, P4, etc.) are based on the original (high variance) variables that are uncorrelated with the previously defined components. Since each additional principal component has a lower variance than the previous one, most of the variance in the sample data can be accounted for within two or three axes.

In this study, principal component analysis provided a means of separating and grouping the compost samples based on their complex PLFA profiles. It also allowed the identification of PLFAs most significant for the separation of groups.

Eigen equations were generated for each principle component axis by an analysis of all samples collected from the pilot-scale facility. These equations separated groups of samples from each other. Paired group data sets (i.e., samples in group I and II, or in group II and III, or in group I and III) were then subjected to additional principle component analyses to further identify the PLFAs responsible for separation. The relative significance of each PLFA was determined from its eigenvalue.

Results

Pilot-Scale Facility Patterns

The principal component analysis of the pilot-scale facility trials in which actual MSW feedstock and the synthetic MSW feedstocks were used, are shown in Fig. 1. The first two principal components explain 85% of the variance.

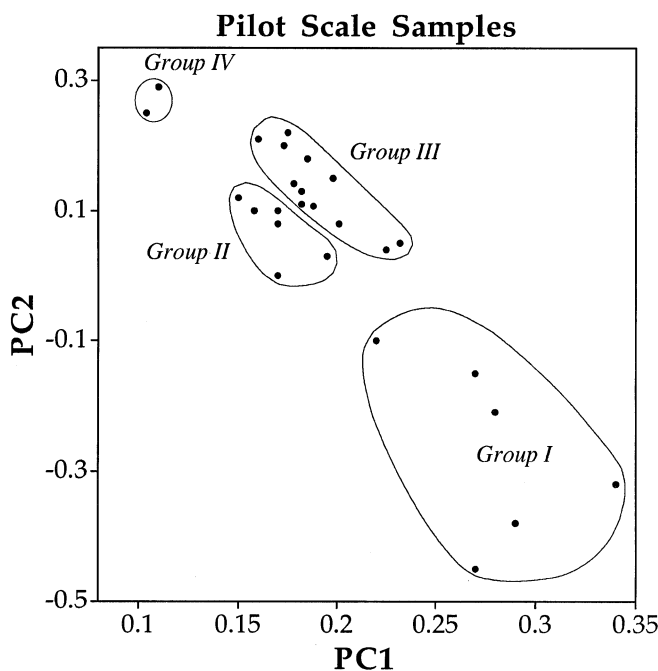


Fig. 1. Principle component analysis of phospholipid fatty acids (PLFA) profiles from a pilot-scale facility for MSW composting. Number of samples = 26. Primary PLFAs for PC1 are cy19:0, i15:0, 16:1 ω 5c, 18:1 ω 7c, i16:0, i17:0, and 18:2 ω 6. Primary PLFAs for PC2 are 18:1 ω 9c, cy19:0, 18:1 ω 7c, i16:0, 18:2 ω 6, 18:3 ω 3, and i15:0.

Table 2. The phospholipids identified as responsible for separation of paired groups (*i.e.*, group I and group II, group II and group III, group I and group III). These were determined by the eigenvalues and eigenequations generated by a principle component analysis of just the paired group samples. Values at the end of a column indicate the temperature range of the composting phase from which group samples were removed

Group I vs. Group II		Group II vs. Group III		Group I vs. Group III	
18:1 ω 9c	i14:0	i16:0	16:1 ω 5c	18:1 ω 9c	i15:0
19:1 ω 6c	i15:0	i17:0	18:1 ω 7c	18:1	a15:0
18:3 ω 3	a15:0	a17:0	18:2 ω 6	19:1 ω 6c	16:1 ω 5c
18:2 ω 6	10me16:0	18:3 ω 3	20:4 ω 6	18:3 ω 3	18:1 ω 7c
20:0	10me18:0	19:1 ω 6c	br19:1	20:0	i15:1
	12me18:0	10me17:0	cy17:0		br19:1
	cy17:0	10me18:0	cy19:0		10me18:0
	cy19:0				cy17:0
	16:1 ω 5c				cy19:0
(35–50°C)	(55–65°C)	(55–65°C)	(20–30°C)	(35–50°C)	(20–30°C)

Within the first three, 92% is accounted for. A scree plot indicated that little more information could be obtained by further additions. Although only two axes are shown in Fig. 1 to represent the data, the three-dimensional separation (PC1, PC2, PC3) was used to identify groups.

The principal component analysis of Fig. 1 yields four major groups. While these groups are based on the lipid profiles of the microbial communities, they separate out by the stage (\approx age) of compost from which they were sampled. Group I consists of samples taken during the initial mesophilic portion, and group II of samples taken during the later thermophilic portion of composting; group III consists of samples taken during the curing phase. Group II and group III were spatially separated along the third principle component axis (PC 3, not shown in Fig. 1). Group IV consists of the two oldest samples (P/MSW/7 and P/SS/7) from compost cured in the 207-liter drums. These two samples can be considered as reference points, reflecting the lipid profile of a relatively mature material. Further analysis was not done on group IV due to the limited number of samples.

Given that the samples in each group can be associated with a particular stage in the composting process, the temperature ranges for each group can also be defined. These ranges and the PLFAs characteristic of each group are presented in Table 2. The defining PLFAs between groups were determined from the eigenvalues generated in an additional principal component analysis of paired groups. Group I is defined by long-chain polyenoics. Group II is dominated by midchain branched lipids and terminally branched lipids, group III by long-chain polyenoics and terminally branched lipids with persistence of the midchain branched lipids. The cyclopropyl lipids (cy17:0 and cy19:0) are higher in the older clusters, as can be seen in the group comparisons. Both

cy17:0 and cy19:0 define the older group in any paired comparison (*i.e.*, I vs. II, II vs. III, or I vs. III).

All samples falling within each group were pooled and averaged to provide the mole percentages of PLFAs along with their standard deviations (Table 3). The cyclopropyl PLFAs, 16:1 ω 5c, 18:1 ω 7c, and i15:0 appear to increase as the composting process proceeds, while the PLFAs 19:1 ω 6c and 18:3 ω 3 decrease. PLFAs that are low during the thermophilic phase (group II), but high in the cooler phases (groups I and III), are 18:1 ω 9c, 18:2 ω 6, and 20:0. PLFAs that are high during the thermophilic phase, but low during cool phases, are i16:0, i17:0, and a17:0.

Comparisons Between Full- and Pilot-Scale Facilities

Fig. 2 shows the results of the principal component analysis of the combined data from the full-scale and pilot-scale samples. In the analysis of the variance, 82% was explained within the first two principal components and 93% by the fourth. A scree plot indicated that little other information could be acquired from further additions.

The trends observed at the pilot-scale level were also evident in this combined data set. The samples that made up group I in Fig. 1 were still found in the lower right of the graph along with the majority of the early stage full-scale facility samples. Samples that made up groups II and III still clustered into the upper left. The early samples from the full-scale composting of nitrogen amended or unamended MSW (F/MSW⁰ and ⁺) fell into the same region as early samples of material composted at the pilot-scale facility. When this amended or unamended MSW was composted at the full-scale and cured at the pilot-scale, it clustered with the other older materials sampled.

Table 3. Mole percentages of significant phospholipids found in groups I, II, and III. Mean mole percentage values are followed by standard deviations. The number of samples were 6, 9, and 8, respectively. The total number of lipids reviewed was 83

PLFA	Group I	Group II	Group III
18:1 ω 9c	26.76 (8.73)	12.13 (3.02)	22.56 (3.76)
19:1 ω 6c	2.04 (1.14)	0.67 (0.45)	0.10 (0.09)
18:3 ω 3	12.95 (9.46)	5.71 (2.08)	0.68 (0.38)
18:2 ω 6	5.68 (8.41)	3.23 (1.92)	14.81 (1.90)
20:0	0.47 (0.10)	0.35 (0.14)	0.43 (0.09)
i14:0	0.23 (0.19)	1.14 (0.33)	1.26 (0.21)
i15:0	1.28 (0.99)	6.43 (0.95)	9.84 (0.92)
a15:0	1.20 (0.72)	3.79 (0.91)	4.94 (0.66)
10me16:0	0.04 (0.08)	0.97 (0.47)	0.89 (0.56)
10me18:0	0.11 (0.16)	1.80 (0.74)	1.89 (0.43)
12me18:0	0.01 (0.01)	0.44 (0.25)	0.83 (0.40)
cy17:0	0.17 (0.24)	1.57 (0.48)	3.80 (0.69)
cy19:0	0.40 (0.31)	6.08 (2.58)	18.84 (2.38)
16:1 ω 5c	0.02 (0.04)	2.61 (3.12)	8.94 (2.08)
i16:0	4.04 (3.67)	13.17 (2.97)	9.12 (1.15)
i17:0	1.75 (1.70)	6.32 (1.40)	5.88 (1.05)
a17:0	2.55 (1.87)	7.09 (1.84)	5.35 (1.21)
10me17:0	0.03 (0.04)	0.94 (0.96)	1.15 (0.63)
18:1 ω 7c	3.05 (1.20)	5.27 (1.52)	13.27 (1.36)
20:4 ω 6	0.40 (0.64)	0.47 (0.25)	1.66 (0.41)
br19:1	0.24 (0.41)	0.32 (0.25)	1.98 (0.47)

Several additional samples were considered in the analysis: a commercially available cow manure compost, a very old full-scale compost (F/MSW/old), and another old pilot-scale compost (P/MSW/old). These were included as indicators of material maturity and stability. Their location in the upper left of the figure with the older F and P/MSW samples suggests that our interpretation of the observed patterns are correct; early stage compost samples fall in the bottom right to midgraph, while later stage and mature ones are found in the top left.

All the samples taken from the composting of the first synthetic formulation (P/S) are also located in the upper left of the figure. Samples of this synthetic MSW were taken across all stages of the composting process, but do not appear to separate out by age. This material had a high N content (relative to C) and its temperature elevation was quick and short. Because of this, the composting of the first synthetic material is not representative of the patterns of lipids observed at the different stages of composting for actual MSW (full- and pilot-scale) or even the second synthetic formulation.

An incremental cluster analysis of the full-scale and pilot-scale samples (Fig. 3) was also performed. The tree produced indicated branching according to the system (full or pilot)

and, to a lesser extent, the age of the compost. The first major division is largely associated with the system where the material was processed. Within the division (primarily made up of samples composted at the full-scale facility), further separations can be made. The **A** cluster consists of full-scale samples drawn during poor aeration of the system; the **B** cluster of samples taken from a better aerated full-scale system. Within **B**, nitrogen addition to the full-scale samples made no noticeable difference. For example, replicate samples of F/MSW⁺/4 and F/MSW⁰/3 fell in the 94% similarity range.

The other half of the first primary division are the samples composted at the Proctor and Gamble pilot facility. The additional separations that occurred within this division appear to be related to the material composted. Cluster **C** consists of early sample dates of actual MSW and synthetic composts. Cluster **D** consists of synthetic MSW samples. This cluster has an age relation; part 1 consists of intermediate aged samples, and part 2 includes older stabilized samples. Cluster **E** is composed of stabilized real MSW samples cured at the pilot-scale facility along with the second synthetic material.

Although the age of the compost is a factor in this cluster

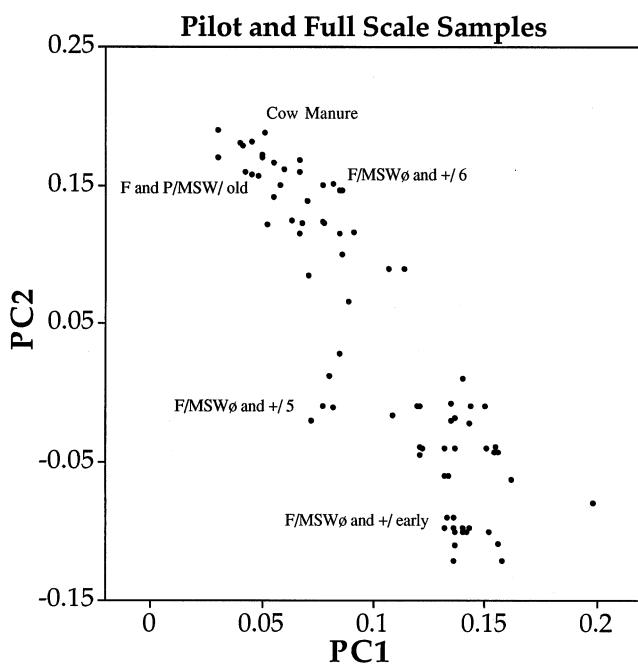


Fig. 2. Principle component analysis of PLFA profiles from both a pilot- and full-scale composting facility. Number of samples = 83. Primary PLFAs for PC1 are 18:1 ω 7c, 18:2 ω 6, 18:3 ω 3, and 18:1 ω 9c. Primary PLFAs for PC2 are cy19:0, i16:0, i15:0, 16:1 ω 5c, 18:1 ω 7c, i17:0, and a17:0.

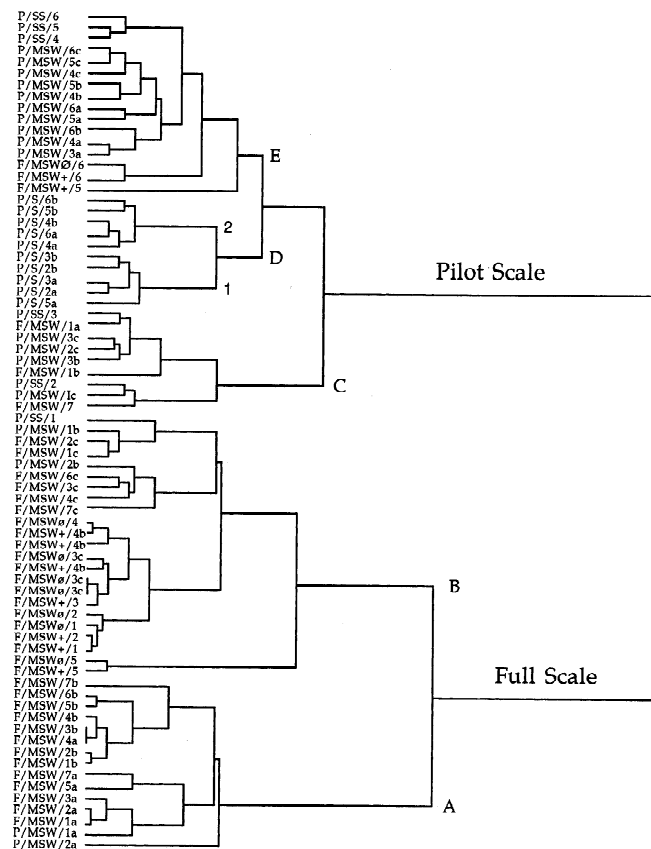


Fig. 3. Incremental cluster analysis of pilot- and full-scale PLFA profiles. Number of samples = 73, replicates cluster at better than 95% similarity.

analysis (e.g., early pilot-scale samples are found in A and B, late samples from full-scale runs are found in E), the principal component analysis better separates the samples on the basis of age.

Discussion

Pilot-Scale Facility Patterns

From the principal component analysis (Fig. 1), four groups were established; the levels of PLFAs within three of these groups were then compared (Table 3). Further analysis allowed us to define the PLFA profiles that characterized each group and distinguished one from another (Table 2). Closer inspection indicated that any given group (and therefore its characteristic PLFAs) was composed of samples that had achieved a specific stage of the composting process. Since PLFA profiles are characteristic of certain microorganisms, the presence or absence of these specific profiles was then used to define the microbial community found in the various stages of composting. The PLFA profile of a given sample, therefore, could potentially be used to define the

stage of composting from which it was taken. This would allow a real-time evaluation of compost processing and the determination of material maturity.

In group I (samples taken from the mesophilic phase of composting) the data points do not group tightly. This is likely due to the high diversity of microorganisms present in the waste stream prior to entering the composting facility [8]. The variance in the data points diminishes in group II, however, as elevated temperatures likely cause the disappearance of many mesophilic microorganisms and, thus, their PLFA markers [18, 26]. The sample points of group III are also close in space. These samples (taken during the curing phase) may represent a microbial community restricted by the type of substrate available, since the material consists mostly of cellulose and lignocellulosic compounds, and/or by their ability to recolonize [11, 19].

In the initial, mesophilic, stage of composting, the dominant PLFAs are 18:1 ω 9c, 19:1 ω 6c, 18:3 ω 3, 18:2 ω 6 and 20:0. Long-chain polyenoics in the 18, 19, and 20 carbon chain length and in particular 18:2 ω 6 indicate a community in which either fungi or algae are dominant [4, 15, 20, 28]. However, in the absence of light or the phototrophic marker, 16:1 ω 13t, these fatty acids are likely fungal in origin. Fungi and bacteria have been implicated as the portions of the microbial community responsible for the initial temperature rise associated with the decomposition of organic residues [11, 13, 21, 24].

During the subsequent thermophilic stage of the composting process, the PLFAs showing increased dominance are i14:0, i15:0, a15:0, i16:0, i17:0, a17:0, 10me16:0, 10me18:0, and 10me17:0. These signature PLFAs are characteristic of actinomycetes and thermophilic bacteria using the aerobic saturase pathway (*Bacillus*, *Thermonospora* and *Micropolyspora*) [2, 4, 14, 17]. All of these types of microorganisms have been isolated from composts during high temperature phases [21, 26]. The strong increase in 15- and 17-carbon chains is a good indication of the presence of thermophilic *Bacillus* sp. [17].

The final, or curing, stage of the composting process is defined by the persistence of actinomycetes markers 10me18:0, 10me16:0, and 10me17:0, and the strong reappearance of the fungal markers 18:2 ω 6 and 18:1 ω 9c [4, 25]. Persistence of actinomycetes into the curing stage, and the reappearance of the fungal community as temperatures drop, has been noted by other researchers [6, 11, 18, 21]. Another indication of lower temperature is revealed by the presence of 20:4 ω 6, a fatty acid that has been associated with protozoans [4].

In addition to reflecting the composition of the microbial community, the results of this study suggest that specific PLFAs might be useful as general indicators of compost maturity. The lipids of cy17:0 and cy19:0 demonstrated a consistent increase as the compost aged (Tables 2 and 3), as did the ratios of cy17:0 to 16:1 ω 7c and 19:0 to 18:1 ω 7c. The cyclopropyl fatty acids have been demonstrated as indicators of metabolic stress, such as a stationary phase of growth and anaerobiosis [10]. Given the frequency of aeration during pilot-scale composting, the indication of anaerobiosis is not as likely as is stationary growth in an aged material. The increase in the amount of these PLFAs should be associated with a decrease in metabolic activity as the readily degradable fraction of the compost is depleted. Therefore, the presence and level of phospholipids cy17:0 and cy19:0 may provide a means of determining the age and stability of a composted material.

Comparisons Between Full and Pilot-Scale Facilities

Compost processed at the full-scale facility, and the composts included as examples of very old material (Fig. 2), showed principal component patterns similar to that of the pilot-scale (Fig. 1). This larger, more inclusive data set appears to reinforce the concept that PLFA profiles reflect the stage of the composting process.

While the trends were the same between samples taken from the full- and pilot-scale facilities, one important difference was noted. Samples from the full-scale facility showed little maturation from the time the material entered the composting bay until it exited. The particular full-scale facility used in this study had difficulty controlling aeration and temperature parameters. Temperatures recorded from some of these sample locations were in the range of 70 to 83°C, levels that are generally detrimental to the microbial community. By impacting the microbial community, these factors seem to have retarded the progression of the composting process in the trench. This problem can be illustrated by following the full-scale nitrogen-amended or unamended samples (F/MSW^{0 and +}). Samples, before and including F/MSW^{0 and +}/4, clustered with the other immature pilot- and full-scale samples (Fig. 2, lower right). In this case, F/MSW^{0 and +}/4 samples were taken from the end of the Royer trench. Only upon curing do the PLFA profiles in samples of these runs gradually shift toward a mature PLFA signature as demonstrated by F/MSW^{0 and +}/5 and 6. This was not restricted to the nitrogen-amended or unamended runs. The other full-scale samples also appeared to finish

composting on the curing pad. The full-scale samples initially taken from the curing pad fall toward the middle (near F/MSW^{0 and +}/5) rather than the upper left (mature material) corner of Fig. 2. The full-scale facility also had a less diverse PLFA structure than the pilot-scale facility; a fact that might help explain the slow development of the compost within this system.

Conclusions

The use of PLFA analysis provides a method for following general patterns in microbial community composition that occur during composting. In this study, a full and pilot-scale facility were compared. In addition, several different types of material were composted. The patterns of change in community structure (based on PLFA profiles) in material processed at the full-scale facility were similar to those observed at the pilot-scale, although they differed significantly in the time of their occurrence. The microbial community within the pilot-scale system progressed more rapidly through the expected changes than did the full-scale community. Regardless, the PLFA profile pattern that emerged (from the complete process) was consistent, predictable, and in agreement with the microbial communities expected. Given this, the PLFA profile may be a useful tool for determining—in any given run—the stage of composting attained. It might also provide a means of identifying stages where a process parameter or other problem is retarding progress. In this fashion, it may allow optimization of a composting situation. PLFA analysis also indicated that a specific community structure was present in stabilized composts.

Characterization of microbial community PLFA structure of stabilized composts may, thus, provide a method of determining maturity and/or stabilization of varied composts. The extraction and analysis of PLFAs is probably too complicated and/or expensive a method to be employed routinely by commercial compost operations. It may be feasible for compost laboratories such as the P&G facility evaluated here to use this type of analysis for process optimization studies and for the evaluation of material (or waste stream) compostability. The applicability and validity of using a pilot-scale facility was verified by this study, where the P&G system realistically simulated and supported the same general microbial communities found at the full-scale facility.

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References

- Bligh EG, Dyer WJ (1959) A rapid method of total lipid extraction and purification. *Can J Biochem Physiol* 37:911–917
- Brennan PJ (1988) Mycobacterium and other actinomycetes. In: Ratledge C, Wilkinson SG (eds) *Microbial lipids*. Academic Press, New York, pp 203–298
- Cook BD, Bloom PR, Halbach TR (1994) A method for determining the ultimate fate of synthetic chemicals during composting. *Compost Sci Util* 2(1):42–50
- Federle TW (1986) Microbial distribution in soil. In: Megusar F, Gantar M (eds) *Perspectives in microbial ecology*. Slovene Society for Microbiology. Ljubljana, Yugoslavia, pp 493–498
- Findlay RH, Trexler MB, Guckert JB, White DC (1990) Laboratory study of disturbance in marine sediments: response of a microbial community. *Mar Ecol Prog Ser* 62:121–133
- Finstein MS (1992) Composting in the context of municipal solid waste management. In: Mitchell R (ed) *Environmental microbiology*. John Wiley & Sons, Wiley Liss Division, New York, pp 355–374
- Finstein MS, Morris ML (1975) Microbiology of municipal solid waste composting. *Adv Appl Microbiol* 19:113–151
- Gaby NS, Creek LL, Gaby WL (1972) A study of the bacterial ecology of composting and the use of *Proteus* as an indicator organism of solid waste. In Murray ED (ed) *Developments in industrial microbiology*, vol 13. American Institute of Biological Science, Washington, D.C., pp 24–29
- Guckert JB, White DC (1986) 2. Phospholipid, ester-linked fatty acid analysis in microbial ecology. In: Megusar F, Gantar M (eds) *Perspectives in microbial ecology*. Slovene Society for Microbiology. Ljubljana, Yugoslavia, pp 455–459
- Guckert JB, Nold SC, Boston HL, White DC (1992) 1. Periphyton response in an industrial receiving stream: lipid-based physiological stress analysis and pattern recognition of microbial community structure. *Can J Fish Aquat Sci* 49:2579–2587
- Hardy GESTJ, Sivasithamparam K (1989) Microbial, chemical, and physical changes during composting of a eucalyptus (*Eucalyptus calophylla* and *Eucalyptus diversicolor*) bark mix. *Biol Fertil Soils* 8:260–270
- Herrmann RF, Shann JR (1993) Enzyme activities as indicators of compost maturity. *Compost Sci Util* 1(4):54–63
- Kononowa MM (1958) *Die Humusstoffe des Bodens*. Deutscher Verlag der Wissenschaften, Berlin
- Kroppenstedt RM, Goodfellow, M (1992) The family *Thermomonosporaceae*. In: Balows A, Truper HG, Dworkin M, Harder W, Schleifer K (eds) *The prokaryotes*, 2nd ed. Springer-Verlag, New York, pp 1085–1114
- Lösel DM (1988) Fungal lipids. In: Ratledge C, Wilkinson SG (eds) *Microbial lipids*. Academic Press, New York, pp 699–806
- McKinley VL, Federle TW, Vestal JR (1982) Effects of petroleum hydrocarbons on plant litter microbiota in an arctic lake. *Appl Environ Microbiol* 43:129–135
- O'Leary WM, Wilkinson SG (1988) G-positive bacteria. In: Ratledge C, Wilkinson SG (eds) *Microbial lipids*. Academic Press, New York, pp 117–202
- Poincelot RP (1974) A scientific examination of the principals and practices of composting. *Compost Sci* 15:24–31
- Riffaldi RR, Levi-Minzi R, Pera A, De Bertoldi M (1986) Evaluation of compost maturity by chemical and microbial analyses. *Waste Management Res* 4(4):387–396
- Ringelberg DB, Davis JD, Smith GA, Pfiffner SM, Nichols PD, Nickels JS, Henson JM, Wilson JT, Yates M, Kampbell DH, Read HW, Stocksdales TT, White DC (1988) Validation of signature phospholipid fatty acid biomarkers for alkane-utilizing bacteria in soils and subsurface aquifer materials. *FEMS Microbiol Ecol* 62:39–50
- Strom PF (1985) Identification of thermophilic bacteria in solid waste composting. *Appl Environ Microbiol* 50:906–913
- Tunlid A, Baird BH, Trexler MB, Olsson S, Finley RH, Odam G, White DC (1985) 1. Determination of phospholipid ester-linked fatty acids and poly *B*-hydroxybutyrate for the estimation of bacterial biomass and activity in the rhizospheres of the rape plant *Brassica napus* (L.). *Can J Microbiol* 31:1113–1119
- Tunlid A, Hoitnik HAJ, Low C, White DC (1989) 2. Characterization of bacteria that suppress *Rhizoctonia* damping-off in bark compost media by analysis by fatty acid biomarkers. *Appl Environ Microbiol* 55:1368–1374
- Updegraff DM (1972) Microbiological aspects of solid-waste composting. In: Murray ED (ed) *Developments in industrial microbiology*, vol 13. American Institute of Biological Science, Washington, D.C., pp 16–24
- Vestal JR, White DC (1989) Lipid analysis in microbial ecology. *BioScience* 39:535–541
- Waksman SA, Cordon TC, Hupoi N (1939) Influence of temperature upon the microbiological population and decomposition processes in composts of stable manure. *Soil Sci* 47:37–61
- White DC (1983) 1. Analysis of microorganisms in terms of quantity and activity in natural environments. In: Slater JH, Whittenbury R, Wimpenny JWT (eds) *Microbes in their natural environments*. Society for General Microbiology Symposium 34:37–66
- White DC, Bobbie RJ, Nickels JS, Fazio SD, Davis WM (1980) 2. Nonselective biochemical methods for the determination of fungal mass and community structure in estuarine detrital microflora. *Bot Mar* 23:239–250