Soil organic matter alterations by short-term composted coffee pulp waste: evidence from pyrolysis-field ionisation mass spectrometry

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Abstract

This study investigates the impact of compost from coffee pulp waste on the composition of soil organic matter (SOM) under controlled laboratory conditions. Compost samples were collected after the thermophilic, mesophilic and the final phase and an equivalent of 48 t ha^{-1} was added to samples of a tropical Nitisol. The soil-compost-mixtures were incubated aerobically for 14 weeks. At the end of incubation the C_{org} concentrations and the total ion intensities of pyrolysis-field ionization mass spectrometry (Py-FIMS) were larger in compost treatments than in the control. Despite of similar initial organic C_{org} and N_t contents among compost amended treatments, less organic C was mineralized following the application of compost from the thermophilic than from later composting phases. The SOM in the soil that received the "thermophilic" compost contained significantly higher proportions of carbohydrates, heterocyclic/nitrilic N-compounds, peptides, sterols and n-fatty acids than the other amended soils. Temperature-resolved volatilization curves for carbohydrates, N-compounds and peptides showed that the enriched compounds covered a wide range of thermal stability. In conclusion, the compost from the thermophilic phase had the best effect in terms of biologically labile C sequestration and stabilization of native organic C in a clay-rich tropical Nitisol. If confirmed for other composts and soils, this may have also considerable economic implications because the time period of composting can be shortened.

Keywords: *compost, soil organic matter, coffee pulp waste, soil carbon sequestration, molecular composition*

Zusammenfassung

Einfluß kompostierter Kaffeefruchtfleisch-Abfälle auf die organische Bodensubstanz: Untersuchungen mit Pyrolyse-Feldionisation Massenspektrometrie

Es wurde der Einfluss kompostierter Kaffeefruchtfleisch-Abfälle auf die organische Bodensubstanz (OBS) unter Laborbedingungen untersucht. Kompostproben wurden nach der thermophilen, mesophilen und der Reifephase entnommen und mit einem tropischen Nitisol vermischt (48 t ha^{-1}). Diese Mischungen wurden unter Laborbedingungen über 14 Wochen inkubiert. Danach lagen die C_{org}-Gehalte und die Gesamtionenintensitäten bei der Pyrolyse-Feldionisation Massenspektrometrie (Py-FIMS) in den Kompost-Varianten über denen der Kontrolle. Trotz gleicher Anfangs-C_{org}- und N_t-Gehalte innerhalb der Kompost-Varianten wurde aus den Varianten mit Kompost aus der thermophilen Phase weniger C_{org} mineralisiert als aus denjenigen mit länger gereiften Komposten. Die OBS in diesen Varianten enthielt signifikant größere Anteile an Kohlenhydraten, N-Verbindungen (Heterozyklen, Nitrile), Peptiden, Sterolen, und *n*-Alkylfettsäuren als in den anderen Kompost-Varianten. Temperaturkurven für die Freisetzung der erstgenannten Verbindungsklassen deuten auf weite Bereiche der thermischen Stabilität hin. Daraus folgt, dass der Kompost auf der thermophilen Phase den besten Effekt für die C-Sequestrierung in diesem Boden hatte. Wenn das für andere Böden und Komposte bestätigt werden könnte, hätte dies möglicherweise auch ökonomische Konsequenzen infolge Verkürzung der Kompostierungsdauer.

Schlüsselwörter: *Kompost, organische Bodensubstanz, Kaffeefruchtfleisch-Abfall, Kohlenstoffspeicherung, molekulare Zusammensetzung*

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1 Introduction

The application of compost to soil has received much attention as an environmentally favorable strategy to use the increasing amount of organic waste and to improve the soil organic matter (SOM) status of agricultural land (Lal, 2004; Weber et al., 2007; Bustamante et al., 2010). Compost application to increase the quantity and improve the quality of SOM is particularly important in strongly weathered tropical soils to overcome the degradation of SOM and improve the soil carbon (C) sequestration (Zech et al., 1997; Lal, 2002, Rivero et al., 2004; Bruun et al., 2010). Studies done by Leifeld et al. (2002), Spaccini et al. (2009) and Eshetu et al. (2012) clearly showed quantitative increases as well as improvements in the SOM quality following compost application to sandy or loamy soils. The effects of compost on composition and stability of SOM depend on various factors including the quality of compost, compost interactions with microbial enzyme systems and mineral soil surfaces (Zech et al., 1997; González-Vila et al., 1999; Grandy and Neff, 2008; Stewart et al., 2011).

Previous research showed that compost application to soil modifies the chemical, physical and biological properties of SOM (Nardi et al., 2004; Adani et al., 2007; Lima et al., 2009). The addition of compost to soil reduced the mineralization of labile compounds of SOM (Fortuna et al. 2003; Piccolo et al., 2004) and increased the humic acid (HA) over the fulvic acid (FA) proportions (Rivero et al., 2004). The latter results were confirmed by Weber et al. (2007), who also reported a relative increase in HA relative to FA with time in soils amended with urban compost. Moreover, the added compost can lead to significant changes in the composition of SOM and minimize labile C mineralization from the soil thereby enhancing the role of SOM as a sink of organic C (Mondini et al., 2007; Spaccini et al., 2009). On the bases of CO_2 respiration results during incubation Mondini et al. (2007) and Sánchez-Monedero et al. (2010) reported an inverse relationship between the organic C mineralized and the degree of stability of the compost added. They concluded that addition of relatively stable compost always reduces the organic C respiration and, thus, stabilize SOM. However, Sánchez-Mondero et al. (2008) observed that the efficiency of added olive mills composts for soil C conservation was independent of the degree of compost stability. González-Vila et al. (1999) stated that extended composting did not contribute to the accumulation of progressively aromatized humic materials but it led to nitrogen and carbohydrate losses. However, most of these results on compost effects on SOM composition and stabilization are based on general chemical information rather than detailed molecular-level studies of SOM. Furthermore, the fate of composts of different origin and stability in tropical soils is completely unknown.

Effects of different management techniques such as compost application on the long term soil C sequestration are difficult to study because biologically induced fluxes take place in short time periods, e.g., hours and days to months whereas geochemical fluxes may take place over longer time periods such as hundreds to millions of years (Paul and Clark,

1989). Therefore, the popular laboratory incubation experiments which measure microbial activity, organic matter decomposition in amended soils (Bernal et al., 1998; Fernandez et al., 2007) and C sequestration potential of arable soils (Mondini et al., 2007; Sanchez-Monedero et al., 2008; Adani et al., 2007) have limitations in applying the results to field conditions and generalisation. For instance, under field conditions the decomposition and stability of organic C is a function of dynamic, heterogeneous and interactive variables such as the amount and quality of plant material entering the soil, the soil mineralogy and chemistry and other soil properties such as moisture, temperature, vegetation etc. The advantage of controlled laboratory incubation experiments is that these factors can be kept constant and restricted so that effects of one variable (e.g. compost quality) can be determined with more precision.

Coffee pulp waste may have significant adverse effects to the environment (Gezahegne et al., 2011; Negassa et al., 2011), but composted it may be a useful amendment of tropical soils. Therefore, it is important to disclose effects of coffee pulp waste composts to SOM contents, composition and stability. Relevant evidence requires the application of molecular-level sensitive analytical methods such as mass spectrometry. For instance, pyrolysis–field ionization mass spectrometry (Py-FIMS) enabled to describe management induced changes in SOM composition in whole soil samples (Schulten and Hempfling, 1992), to characterize SOM in sewage farm soils (Leinweber et al., 1996), to trace different phases of biowaste composting (Smidt et al., 2005) and to characterise the organic matter in fresh agro-industrial waste including coffee pulp (Negassa et al., 2011). Recently, Eshetu et al. (2012) used Py-FIMS in conjunction with pyrolysis gas chromatography/mass spectrometry to study long-term compost effects on the molecular composition of SOM developed in humus free Pleistocene loamy marl. Most recently we observed that composts taken after the thermophilic phase may have an overall better effect to soil C sequestration than mature composts (Eshetu et al., 2013). However, neither Py-FIMS nor another highly sensitive speciation method has been applied so far to disclose effects of differently aged or stabilised composts from coffee pulp waste on SOM in tropical soils.

Therefore, the objectives of the study were (1) to evaluate the effect of compost age on the molecular structure of SOM in a tropical Nitisol amended with compost derived from different phases of composting of coffee pulp waste, and (2) to investigate which constituents of organic matter were decomposed or relatively enriched, when differently stabilized composts were incubated in the Nitisol sample to simulate the microbial decomposition.

2 Materials and methods

2.1 Soil and composting samples

The soil was sampled at a coffee plantation farm located in the South-Western Highlands of Ethiopia and classified as Nitosol (ISSS-ISRIC-FAO, 1998). The sampling area is situated

36°36'E longitude and 7°56'N latitude. The elevation varies between 1533-1548 m a.s.l. Mean minimum and maximum temperatures range from 11 to 14 °C and 26 to 30 °C, respectively, and the mean of precipitation ranges from 1131 to 1150 mm. Thus, the climate may be indicated as warm tropical. A total of 50 subsamples were taken from 5.91 ha land, traversed in a zig-zag pattern, from soil depth of 0 to 30 cm using a GPS and an auger. The subsamples were well mixed in a plastic container and then a representative sample of 3 kg was placed in a labeled plastic bag. The soil sample was air-dried and milled to pass a 2 mm sieve. Its main characteristics are compiled in Table 1.

Table 1

Main characteristics of soil, fresh waste and differently stabilized compost samples (RCPW: fresh coffee pulp waste, COM: compost, T: thermophilic phase, M: mesophilic phase, F: final stage, C_{org}: total organic carbon, N_t: total nitrogen, S_t: total sulfur, C/N: carbon to nitrogen ratio, EC: electrical conductivity).

Parameter	Soil	RCPW	COM1-T	COM1-M	COM1-F
pH	5.2	5.8	8.8	9.3	9.1
C_{org} (g kg^{-1})	38.6	397.0	151.0	149.0	139.2
N_t (g kg^{-1})	3.2	21.1	10.5	12.7	12.7
S_t (g kg^{-1})	0.8	3.1	1.7	2.2	2.4
C/N	11.9	18.9	14.4	11.7	11.0
EC (mS cm^{-1})	2.4	6.7	2.0	2.1	2.1
Soil texture					
% Sand	1		ND	ND	ND
% Silt	14		ND	ND	ND
% Clay	84		ND	ND	ND
Phytotoxicity (Germination index in %)					
With cress		3	106	109	102
With Radish		26	121	120	125

The compost was produced from 800 kg raw coffee pulp waste (RCPW) in a method of small heap composting. Heaps were piled in a bamboo box of dimensions 1.5 m (l) × 1.0 m (w) × 0.8 m (h)). The platform used has been considered as pilot compost production unit by the Addis Ababa city council environmental protection authority, Ethiopia. The regional climate is tropical, with average temperatures between 15 °C and 25 °C. Piles were constructed for experimental purposes with duplicates. About 5 % (w/w) soil and 10 % (w/w) of garden trimmings (dry leaves, and young tree branches) were added as additional source of microbial colonization and to improve the pile structure. The experiment lasted from November 2010 to February 2011. The moisture content of the compost was manually estimated on site by hand squeezing the compost tightly. Moisture contents above 40 % were indicated by a formation of a ball up on squeezing of the compost. Depending on the situation the piles were watered so as to always maintain > 40 % (w/w) moisture. Samples

were taken once a week for pH and moisture content measurements in the laboratory. Temperatures were measured daily for the first two weeks and in a three-days-interval during the next period always in the early morning using digital thermometers (0.5 and 1.0 m in length) at two different points of the heaps (25 and 60 cm depth). The piles were manually turned each week during the thermophilic phase (45°C to 69 °C) which lasts 30 days, and then in a 15-days-interval during the maturation phase (< 43 °C). Temperature was considered as a major indicator how well the composting process was progressing. The composting was considered to be finished when the temperature of the mixture remained stable and near ambient (about 21 °C) after 16 weeks.

Representative samples (about 1 kg) were taken by mixing nine subsamples from different levels of the section in the pile at different phases of the composting, day 8 (thermophlic phase = COM-T), day 36 (mesophilic phase = COM-M) and day 119 (final compost = COM-F). In this way composts of different stability were obtained. The samples were air dried and ground to pass a 0.5 mm sieve. Main properties are shown in Table 1.

Aerobic incubation of the soil-compost-mixtures was done by mixing 10 g dry weight of sieved soil samples (< 2 mm) with fresh and composted coffee pulp waste at desired application rates (48 t ha^{-1}) and placed in 100 ml incubation vessels in five replicates each. They were monitored as treatment S+RCPW, S+COM1-T, S+COM1-M and S+COM1-F. Soil controls were run without any amendment. Distilled water (1 to 5 ml) was added to the mixtures to keep the moisture at 60 % of water-holding capacity. The incubation was carried out in a temperature-controlled incubator at 25 °C for 98 days. Subsamples (2 g) were taken destructively from each treatment at the end of incubation for elemental and Py-FIMS analyses.

2.2 Chemical analyses: basic data and molecular – chemical characterization

The soil samples were analyzed for pH and EC in 0.01 M CaCl$_2$ suspensions (1:2.5 w/v), and fresh and composed waste samples were analyzed for pH and EC in H$_2$O suspensions (1:10 w/v). The concentrations of total organic carbon (C_{org}), total nitrogen (N_t) and total sulfur (S_t) were determined for soil, compost and amended soil samples using a CNS analyzer (Vario EL III; Elementar Analysensysteme, Hanau, Germany). RCPW and compost phytotoxicity was determined according to the modified method of Zucconi et al. (1985) and described and discussed in Eshetu et al. (2013).

For Py-FIMS about 3 mg of the air dried, ground and homogenized samples were thermally degraded in the ion source (emitter: 4.7 kV, counter electrode -5.5 kV) of a double-focusing Finnigan MAT 95 mass spectrometer. All samples were heated in a vacuum of 10^{-4} Pa from 50 °C to 700 °C, in temperature steps of 10 °C over a time period of 18 minutes. Between magnetic scans the emitter was flash-heated to avoid residues of pyrolysis products. About 65 magnetic scans were recorded for the mass range *m/z* 15 to 900. Ion intensities were referred to 1 mg of a sample. For each of the

single scans, the absolute and relative ion intensities of ten classes of compounds in the organic matter were calculated by summation of the ion intensities of indicator signals to obtain thermograms of their volatilization and averaged Py-FI mass spectra. This procedure was done for each three replicate measurements per soil sample and the results were averaged for statistical analysis. Information on the molecular size distribution of pyrolysis products was obtained by calculating the molecular weight averages M_w (weight-based) and M_n (number-based), and the polydispersity (I = M_w / M_n) (Lattimer and Schulten, 1983). Detailed descriptions of the Py-FIMS methodology are published in Leinweber et al. (2009).

2.3 Statistical analyses

Means were calculated for physicochemical properties of soils, composts, amended soils and ion intensities from Py-FIMS. Comparisons between means of ion intensities of compound classes and polydispersity in different treatment were made by a One Way ANOVA test. Multivariate statistical evaluation of Py-FI mass spectra was done by principal component analysis to test which *m/z* signals contributed to differences among samples. All statistics were computed using data analysis and graphic software (Origin 8.1G).

3 Results and Discussion

3.1 Elemental composition

Organic amendments increased the C_{org} concentrations by 7.8 g kg⁻¹ (RCPW) and about 2 g kg⁻¹ (with the addition of composts) at start of the incubation (Table 2). The increases in N_t concentrations were proportionally. These differences in C_{org} and N_t concentrations originate from the different amounts of organic matter applied with RCPW and the composts (Lima et al., 2009). At the end of incubation all samples had lower C_{org} and N_t (except the S + RCPW) concentrations. The C_{org} losses were larger in the soil amended with RCPW (10.5 %) than in the samples amended with composts (2 % to 4.1 %). This difference in C_{org} losses by respiration are explained by the decomposition of easily degradable organic com-

pounds from the RCPW which in the composts had been either mineralized (Bernal et al., 1998) or otherwise stabilized. Furthermore, the observed smallest C_{org} losses by only 2.0 % in the samples amended with compost from the thermophilic phase disagrees with the findings of (Leifeld et al., 2002; Mondini et al., 2007). These authors found an inverse relationship between the organic C mineralized and the degree of stability of the compost added in a laboratory incubation experiment and concluded that composts require a maturation phase for humification and organic matter stabilization. Explanation of this disagreement requires a detailed molecular-chemical investigation of the different composts and soil-compost-mixtures as described below.

3.2 Pyrolysis – field ionization mass spectrometry (Py-FIMS)

In the Py-FIMS the thermograms of total ion intensity (TII) shows single peaks for all the control soil and the soil amended with RCPW (Figure 1), and similar for the three samples with compost amendment (Figure 2). Addition of the organic materials slightly changed the peak temperatures of maximum volatilization of organic molecules from about 500 °C in the control to 470 to 500 °C in the samples with organic amendments (see Figure 1 and compare with Figure 2). Furthermore, the pronounced shoulder at 420 °C seen at the control soil sample (Figure 1) disappeared in the samples with organic amendments (Figure 2), indicating some stabilization of labile organic matter by the organic amendments. The signal patterns of the Py-FI mass spectra revealed differences among compost samples originating from the different duration of the composting. For instance the prominent signals at *m/z* 58, 72, 96, and 110 indicate a particularly high relative intensity of carbohydrates in sample S+COM1-T (Figure 2). The very intense *m/z* 84 in this treatment can be explained by pyrolysis products of carbohydrates and peptides. Furthermore, *m/z* 67 (pyrrole) and *m/z* 117 (indole) seem to be more abundant in this than in the other composts. Moreover, the signals with highest molecular masses originating from sterols and long-chained *n*-fatty acids (e.g., *m/z* 390, 424) were detected in this treatment only (Figure 2). On the other hand, intense signals of phenols/lignin

Table 2

Elemental composition (g kg⁻¹) of control soil sample, and soil samples amended with row coffee pulp waste (S+RCPW) and composts from coffee pulp waste taken at the thermophilic (S+COM1-T), mesophilic (S+COM1-M) and final maturation (S+COM1-F) phase of composting at start (Day 1) and end of an aerobic incubation (Day 98). The right column indicates the relative decrease in the C_{org} concentrations during the incubation.

Treatments	Day 1				Day 98				
	C_{org}	N_t	S_t	C/N	C_{org}	N_t	S_t	C/N	C_{org} loss (%)
Control	39.8	3.3	0.7	12.2	38.3	3.2	0.7	11.8	3.7
S+RCPW	47.6	3.7	0.8	12.9	42.6	3.7	0.8	11.6	10.5
S+COM1-T	42.0	3.5	0.7	11.9	41.2	3.4	0.8	11.9	2.0
S+COM 1-M	42.0	3.6	0.7	11.6	40.3	3.4	0.7	11.9	4.1
S+COM 1-F	41.8	3.6	0.8	11.7	40.7	3.4	0.7	12.0	2.6

monomers *(m/z* 168, 180, 194), lipids *(m/z* 202, 216, 230), homologous series of alkanes *(m/z* 252, 266, 280, 294, 308) and alkylaromatics *(m/z* 218) were found in all spectra.

A more detailed spectra evaluation by PCA indicated that 92.4 % of the total variance in the whole data set from the three soil-compost-mixtures could be explained by the first two principal components (not shown). For 34 *m/z* signals the univatiate wilks's lambda was significant at a level of p < 0.01. The signals with the highest discriminating powers among treatments S+COM1-F, S+COM1-M and S+COM1-T are *m/z* 114, 71, 176, 434, 399, 113, 85, 177, 189, 468, 157, 84, 219, 83, 69, 253, 228, 143, 44, 46, 60, 221, 252, 202, 141, 72, 53, 269, 107, 82, 200, 88, 152, 233, 443 and 167 (arranged with decreasing importance for discrimination). For instance the pentose marker *m/z* 114 from plant material was significantly higher in treatment S+COM1-T but showed no significant difference between composts from the mesophilic and final phase. On the other side, the relative proportions of the lignin marker *m/z* 178 (coniferyl aldehyde) (Leinweber et al., 2009) was significantly higher in S+COM1-M and S+COM1-F than in S+COM1-T. This provides unequivocal evidence that duration of the composting has an effect on the molecular composition not only of the compost (Smidt et al., 2005) but also of a soil amended with this compost.

The data derived from the Py-FI mass spectra pointed to similarities but also significant to differences among samples at the end of the incubation (Table 3). The proportions of volatile matter, ranging from 20 to 25 %, differed insignificantly, indicating that organic matter and adsorbed water were not greatly affected by the soil amendments. The TII values were significantly larger in all amended soils than in the control without significant differences (p < 0.05) among the amended soil samples. Assignment of markers signals to important compound classes of SOM revealed significantly larger intensities of carbohydrates, peptides and sterols in S+COM1-T than in the other treatments. Difference were largest

between S+COM1-T and the control accounting for 4.4×10^6 counts mg^{-1} for carbohydrates, 3.9×10^6 counts mg^{-1} for peptides and (0.2×10^6 counts mg^{-1} for sterols (Table 3). The ion intensities of phenols/lignin monomers, alkylaromatics, and N-compounds were significantly lower in the control than in the amended soils but did not significantly differ among the amendments.

The relative ion intensities of compound classes indicated that carbohydrates (12.8 %) and peptides (12.2 %) were the major SOM constituents in S+COM1-T (Table 3, see values in parentheses). Furthermore, this sample contained significantly more N-compounds (7.1 %) and sterols (0.6 %) than any other sample. Phenols/lignin monomers (13.1 to14.0 %) and alkylaromatics (14.9 to 15.4 %) were more abundant in the other amended soil samples and the control. Suberin was the compound class with smallest proportions in all samples.

The molecular weight averages M_n (142 g mol^{-1}) and M_w (194 g mol^{-1}) of S+COM1-T were significantly lower than those of the other treatments (M_n: 171 to 175 g mol^{-1}; M_w: 205 to 207 g mol^{-1}). This was due to the dominance of low molecular-weight organic compounds such as carbohydrates and N-compounds. However, a significantly higher M_w/M_n ratio (1.4) in treatment S+COM1-T indicated a higher polydispersity of the SOM in this treatment compared to the other organic amendments (1.2). This was due to the fact that the SOM in treatment S+COM1-T are complex mixtures of low molecular mass compounds (such as carbohydrates) to high molecular masses compounds (such as *n*-fatty acids and sterols) (Figure 1, Table 3).

The TII larger in the compost fertilized than in other treatments (Table 3) agreed with the C_{org} concentrations (Table 2). This confirms previous studies in which the TII from Py-FIMS also corresponded to the C_{org} concentrations of different agricultural soils (Schulten and Hempfling, 1992), organic waste materials (Franke et al., 2006) and compost-amended soils (Eshetu et al., 2012). The results provide evidence for an

Table 3

Results of pyrolysis-field ionization mass spectrometry: volatile matter (VM, %), total ion intensity (TII, 10^6 counts mg^{-1}), ion intensities for 10 important compound classes of organic matter (CARB: carbohydrates, PHLM: phenols/lignin monomers, LIPID: lipids, NCOMP: N-compounds, ALKY: alkylaromatics, LDIM: lignin dimers, STER: sterols, PEPT: peptides, SUBE: suberin, FATTY: *n*-C$_{16}$ to *n*-C$_{34}$ fatty acids in 10^6 counts mg^{-1}), relative ion intensity (values in parenthesis, % TII) and molecular weight averages (g mol^{-1}). For explanation of treatments see Table 1). Values followed by the same letters with in a column and column without letters were not significantly different at the 5 % probability level.

Treatment	VM	TII	CARB	PHLM	LDIM	LIPID	ALKY	NCOMP	STER	PEPT	SUBE	FATTY	M_n	Mw	Mw/Mn
CONTROL	23.3a	30.6b	2.4 (7.7)a	4.3 (14.0)a	2.7 (8.8)ab	1.1 (3.4)a	4.6 (15.0)a	1.8 (5.8)a	0.1 (0.2)a	2.6 (8.3) b	0.0 (0.0)	0.0 (0.1)a	175.7a	206.5a	1.2a
S + RCPW	21.1a	46.4a	3.6 (7.7)a	6.1 (13.1)b	3.9 (8.3)ab	1.5 (3.3)ab	7.1 (15.2)b	2.6 (5.7)b	0.1 (0.1)a	3.6 (7.8)ab	0.0 (0.0)	0.0 (0.0)a	178.5a	207.3a	1.2a
S + COM1-T	20.1a	53.3a	6.8 (12.8)b	5.8 (11.0)ab	2.6 (5.0)a	1.0 (1.9)a	5.9 (11.1)b	3.8 (7.1)c	0.3 (0.6)b	6.5 (12.2)c	0.0 (0.0)	0.1 (0.1)b	141.5b	194.2b	1.4b
S + COM1-M	22.1a	54.3a	4.2 (7.7)a	7.2 (13.2)b	4.4 (8.1)b	1.6 (3.0)b	8.1 (14.9)b	3.2 (6.0)bc	0.1 (0.2)a	4.5 (8.2)a	0.0 (0.0)	0.0 (0.1)ab	171.6a	204.2a	1.2a
S + COM1-F	25.5a	46.6a	3.9 (8.5)a	6.4 (13.8)b	3.4 (7.4)ab	1.3 (2.8)ab	7.2 (15.4)b	2.7 (5.8)ab	0.1 (0.2) a	4.0 (8.5)a	0.0 (0.0)	0.0 (0.1)ab	174.4a	205.7a	1.2a
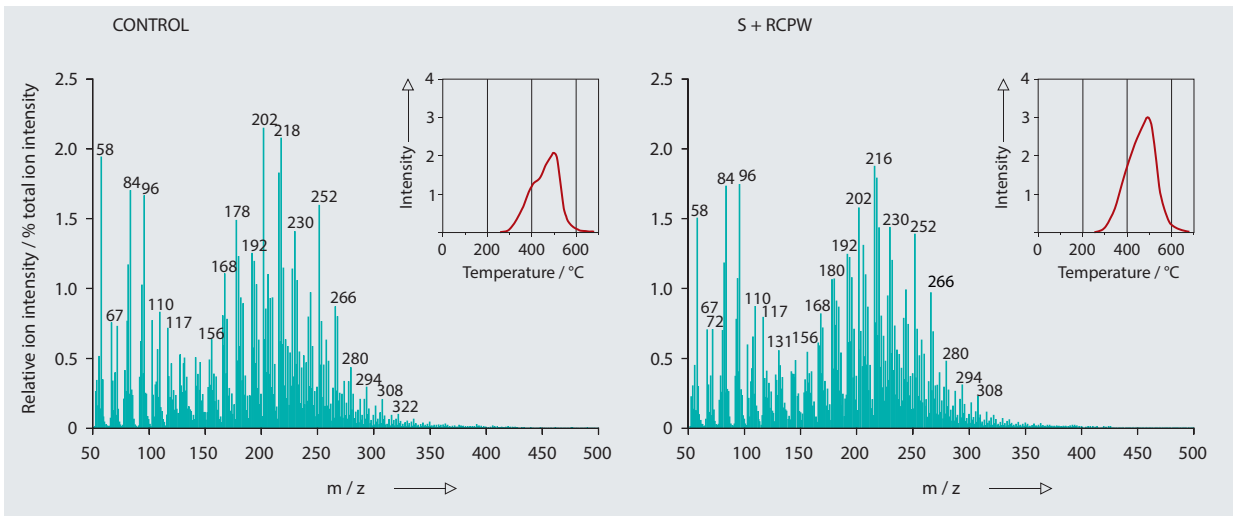
Figure 1

Summed and averaged pyrolysis-field ionization mass spectra and thermograms of total ion intensity (TII, upper right inserts) of a Nitisol soil sample amended with fresh coffee pulp waste (S+RCPW) and the non-amended control sample of the Nitisol

exogenous C input to these treatments from compost and microbial biomass (Leinweber and Reuter, 1992; Spaccini et al., 2009; Eshetu et al., 2012).

The signal patterns (Figure 2) and summed intensities of compound classes (Table 3) indicated the clearest impact of compost from the thermophilic phase on the SOM composition. The particularly high ion intensities of carbohydrates, peptides and heterocyclic N compounds are explained by their enrichment in the thermophilic phase of composting. Obviously, these compounds were not mineralized when the "thermophilic compost" is incubated with the Nitisol sample. This resistance of otherwise labile compounds (carbohydrates and peptides) can be explained by stabilization at reactive surfaces (Gregorich et al., 1996; Zech et al., 1997; Grandy and Neff, 2008; Bruun et al., 2010; Bustamante et al., 2010) of the clay-rich (84 % < 2 µm) Nitisol. The abundance of reactive clay surfaces is crucial, because both carbohydrates and N-compounds were completely decomposed in re-circulated leachates from organic waste in large-scale lysimeters that offered no contact with reactive surfaces (Franke et al., 2007). Unlike the high content of *n*-fatty acids and sterols in the raw coffee pulp waste (Negassa et al., 2011) signals of these compounds were detected only in treatment S+COM1-T. This can be explained by the decomposition of these compounds in the later stages of compositing. Smidt et al. (2005) also reported that fatty acids and sterols changed most during composting of yard and kitchen wastes studied by Py-FIMS and FT-IR spectroscopy.

The similar intensities of phenols/lignin monomers, lignin dimers, lipids and alkylaromatics among the compost-amended soils revealed that these compound classes were little affected by the degree of stabilization of composts and subsequent short-term incubation. However, their higher contents in amended soils compared to the control indicate enrichments of non-decomposable stable compounds by

compost addition, irrespective of the compost maturation (Buurman et al., 2007; Stewart et al., 2011). For instance similarly large proportions of phenols/lignin monomers, lipids and alkylaromatics were also observed in both fresh and decomposed plant materials (Leinweber and Schulten, 1999), in a fresh coffee pulp waste (Negassa et al., 2011), and in long-term compost-fertilized soils (Eshetu et al., 2012). The small suberin proportions in all samples (Table 3) are explained by the origin of suberin from bark and outer covering of twigs (Hempfling et al., 1991) which were absent in these samples. The average molecular weights of the compost-amended soils agree with the results recorded for SOM accumulated after 19 and 34 years of a humus-formation experiment (Schulten et al., 1992), are greater than values observed in soils under potato cultivation and crop rotation (Schulten and Hempfling, 1992) and are below values from pig slurry fractions (Aust et al., 2009).

Figure 3 shows the temperature-resolved volatilization curves of those compound classes that were particularly enriched in the soil incubated with compost from the thermophilic phase. The maximum volatilization occurred at 390 °C (carbohydrates), 420 °C (N-compounds) and 410 °C (peptides). The thermograms indicate that compounds enriched in treatment S+COM1-T were volatilized over the whole temperature range. In other words, there is no enrichment in a particularly low or high temperature range. This was also true for the comparison of the other organic amendments and the control (Figure 3).

The thermograms for N-compounds and peptides in Figure 3 show some differences to the corresponding thermograms obtained for similarly treated compost from coffee pulp waste and fruit waste (Eshetu et al., 2013). In the latter more complex compost the volatilization maxima were at 10 to 50 °C higher pyrolysis temperature, indicating that the organic parent materials affect the stability of the compost in
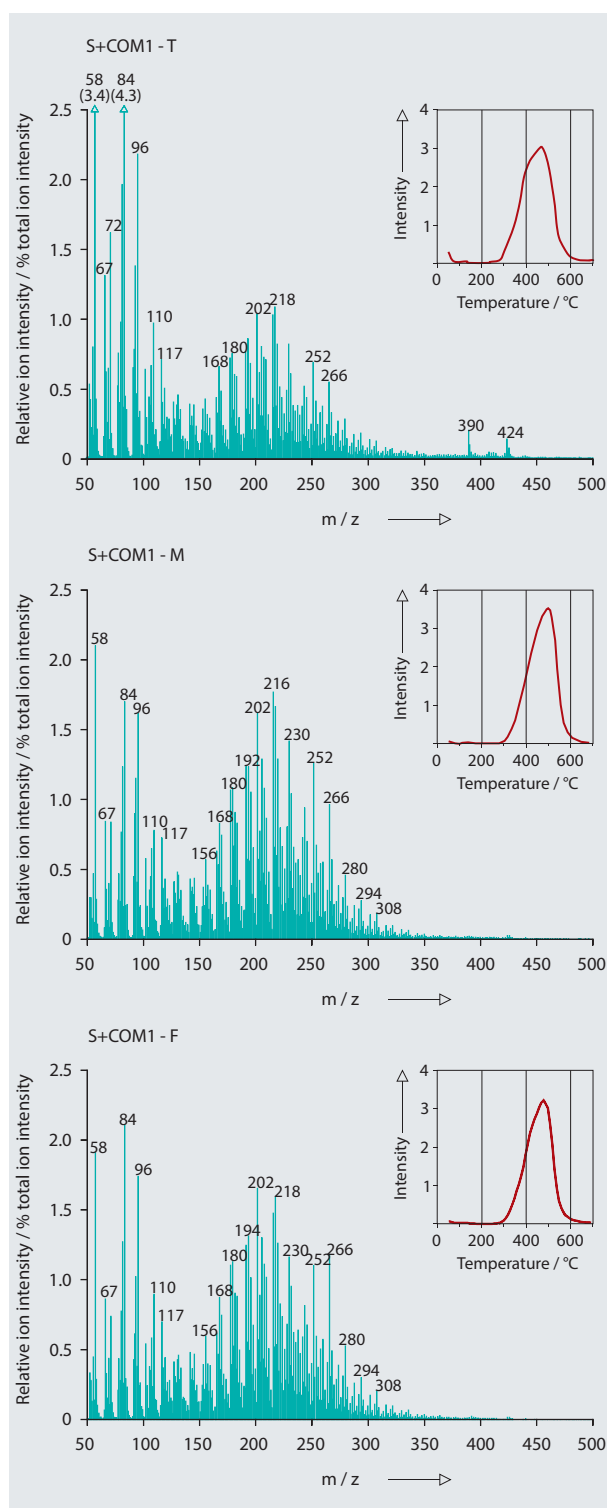
Figure 2

Summed and averaged pyrolysis-field ionization mass spectra and thermograms of total ion intensity (TII, upper right inserts) of Nitisol soil samples amended with composts taken in different phases of composting (COM – compost; T – thermophilic phase; M – mesophilic phase; F – final stage)
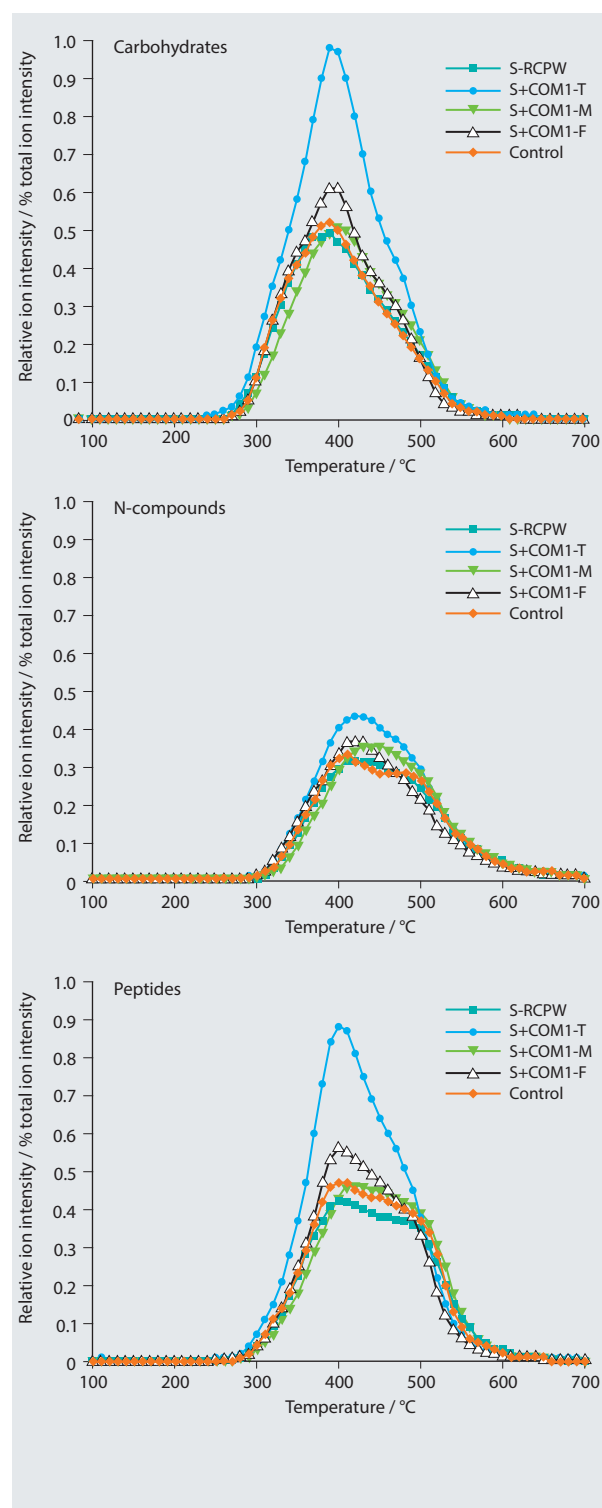
Figure 3

Thermograms for the evolution of carbohydrates, N-compounds, and peptides derived from the pyrolysis-field ionization mass spectra

soil even when it was removed from the composting in an early phase. The broad temperature range in which the enriched compound classes were thermally volatilized point to the presence of both, labile and rather stabile humified organics which originate from microbes and the compost (Schulten and Hempfling, 1992; Stuczynski et al., 1997; Nierop et al., 2001; Kögel-Knabner, 2002). The lowest proportion of these fractions in S+RCPW provides evidence that non-altered plant materials scarcely contributed to the observed enrichment when the compost COM1-T was incubated with soil. More likely is the origin of these carbohydrates and protein from the microbial biomass such as fungi and bacteria (Zech et al., 1997) which possibly reach a maximum abundance in the thermophilic phase of composting. Furthermore, the described enrichment in carbohydrates and protein-derived compounds well agrees with findings from Lima et al. (2009) who reported larger proportions of these compound classes in compost-amended soils than in soil that received fresh manure. Their soil was dominated by the silt fraction (76 %) and compost effects were determined by ^{13}C NMR spectroscopy. Thus, the abundance of reactive surfaces from silt (Lima et al., 2009) or clay (Nitisol of the present study) supports the stabilization of these labile organic compounds of microbial origin, making them non-decomposable in short-term incubation under optimized aerobic and moisture conditions. The observed lower proportions of carbohydrates, peptides and N-compounds in the soil amended with the raw coffee pulp waste or composts taken at later stages of composting can be explained by the decrease in microbial activity and microbial residues after the rapid consumption of easily degradable organic matter and high temperatures during the thermophilic phase (Dignac et al., 2005) and a progressive decrease of carbohydrate content during composting (Chefetz et al., 1996; Spaccini and Piccolo, 2007).

4 Conclusions

The Py-FIMS for the first time provided compelling evidence that the phase after which compost is applied to soil finally determines how stable it is in the soil. Furthermore, the results from this study suggest that it may be better to end up the composting of coffee pulp waste immediately after a short thermophilc phase and apply this "fresh" compost to soil rather than processing the compost through a mesophilic (cooling) and maturation phase of composting. Since the Nitisol used in this study was particularly rich in clay, binding of intermediate decomposition products and microbial metabolites to reactive surfaces or in clay aggregates is the best explanation for the enrichment of carbohydrates and various non-cyclic and cyclic N-containing compounds. However, our results based on small-scale laboratory incubation experiment are restricted to a single soil type and under exclusion of factors such as temperature/moisture fluctuations, meso- and macrofauna or plants and other field heterogeneity in general, so that further research in the field or using field replicates as well as investigating different soil types is recommended.

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References

Adani F, Genevini P, Ricca G, Tambone F, Montoneri E (2007) Modification of soil humic matter after 4 years of compost application. Waste Manag 27:319-324

Aust M-O, Thiele-Bruhn S, Eckhardt K-U, Leinweber P (2009) Composition of organic matter in particle size fractionated pig slurry. Biores Technol 100:5736-5743

Bernal MP, Sánchez-Monedero MA, Paredes C, Roig A (1998) Carbon mineralization from organic wastes at different composting stages during incubation with soil. Agric Ecosyst Environ 69:175-189

Bolan NS, Kunhikrishnan A, Choppala GK, Thangarajan R, Chung JW (2012) Stabilization of carbon in composts and biochars in relation to carbon sequestration and soil fertility. Sci Total Environ 1(424):264-270

Bruun TB, Elberling B, Christensen BT (2010) Lability of soil organic carbon in tropi cal soils with different clay minerals. Soil Biol Biochem 42:888-895

Bustamante MA, Said-Pullicino D, Paredes C, Cecilia JA, Moral R (2010) Influences of winery–distillery waste compost stability and soil type on soil carbon dynamics in amended soils. Waste Manag 30(10):1966-1975

Buurman P, Peterse F, Martin GA (2007) Soil organic matter chemistry in allophanic soils : a pyrolysis-GC/MS study of a Costa Rican Andosol catena. Eur J Soil Sci 58:1330-1347

Chefetz B, Hatcher PG, Hadar Y, Chen Y (1996) Chemical and biological characterization of organic matter during composting of municipal solid waste. J Environ Quality 25:776-785

Dignac MF, Houot S, Derenne S, Francou C, Derenne S (2005) Pyrolytic study of compost and waste organic matter. Org Geochem 36:1054-1071

Eshetu B, Jandl G, Leinweber P (2012) Compost changed soil organic matter molecular composition : a study by Py-GC/MS and Py-FIMS. Compost Sci Util 20:230-238

Eshetu B, Baum C, Leinweber P (2013) Compost of different stability affects the molecular composition and mineralization of soil organic matter. Open J Soil Sci 3:58-69

Fernández JM, Hernández D, Plaza C, Polo A (2007) Organic matter in degraded agricultural soil amended with composted and thermally-dried sewage sludge. Sci Total Environ 378:75-80

Fortuna A, Harwood RR, Kizilkaya K, Paul EA (2003) Optimizing nutrient availability and potential carbon sequestration in an agro-ecosystem. Soil Biol Biochem 35:1005-1013

Franke M, Jandl G, Leinweber P (2006) Organic compounds in re-circulated leachates of aerobic biological treated municipal solid waste. Biodegradation 17:473-485

Franke M, Jandl G, Leinweber P (2007) Analytical pyrolysis of re-circulated leachates : towards an improved municipal waste treatment. J Anal Appl Pyrolysis 79:16-23

Gezahegne B, Fikre L, Mulatu W (2011) Exploring the suitability of coffee pulp compost as growth media substitute in greenhouse production. Int J Agric Res 6(3):255-267

Gleixner G, Poirier N, Bol R, Balesdent J (2002) Molecular dynamics of organic matter in a cultivated soil. Org Geochem 33:357-366

González-Vila FJ, Almendros G, Madrid F (1999) Molecular alterations of organic fractions from urban waste in the course of composting and their further transformation in amended soil. Sci Total Environ 236:215-229

Grandy AS, Neff JC (2008) Molecular C dynamics downstream : the biochemi-
cal decomposition sequence and its impact on soil organic matter struc-
ture and function. Sci Total Environ 404:297-307

Gregorich EG, Monreal CM, Schnitzer M, Schulten H-R (1996) Transformation
of plant residues into SOM : chemical characterization of plant tissue, iso-
lated soil fractions, and whole soils. Soil Sci 161:680-693

Hempfling R, Simmleit N, Schulten H-R (1991) Characterization and chemo-
dynamics of plant constituents during maturation, senescence and
humus genesis in spruce ecosystems. Biogeochemistry 13:27-60

ISSS-ISRIC-FAO (1998) World Reference Base for Soil Resources. FAO, World
Soil Resources Report No. 84, Rome

Kögel-Knabner I (2002) The macromolecular organic composition of plant
and microbial residues as inputs to soil organic matter. Soil Biol Biochem
34:139-162

Lal R (2002) The potential of soils of the tropics to sequester carbon and miti-
gate the greenhouse effect. Adv Agron 74:155-192

Lal R (2004) Soil carbon sequestration impacts on global climate change and
food security. Science 304:1623-1627

Lattimer RP, Schulten H-R (1983) Field desorption of hydrocarbon polymers.
Int J Mass Spectrom Ion Phys 52:105-116

Leifeld J, Siebert S, Kogel-Knaber I (2002) Changes in the chemical composi-
tion of soil organic matter after application of compost. Eur J Soil Sci
53:299-309

Leinweber P, Blumenstein O, Schulten H-R (1996) Organic matter composition
in sewage farm soils : investigations by ^{13}C-NMR and pyrolysis-field ioniza-
tion mass spectrometry. Eur J Soil Sci 47:71-80

Leinweber P, Jandl G, Eckhardt K-U, Schlichting A, Hofmann D, Schulten H-R
(2009) Analytical pyrolysis and soft ionization mass spectrometry. In:
Senesi N, Xing B, Huang PM (eds) Biophysico-chemical processes
involving natural nonliving organic matter in environmental systems.
Hoboken : Wiley, pp 533-582

Leinweber P, Schulten H-R (1993) Dynamics of soil organic matter studied by
pyrolysis-field ionization mass spectrometry. J Anal Appl Pyrolysis 25:
123-136

Leinweber P, Reuter G (1992) The influence of different organic fertilization
practices on concentrations of organic carbon and total nitrogen in par-
ticle-size fractions during 34 years of a soil formation experiment in
loamy marl. Biol Fertil Soils 13:119-124

Leinweber P, Schulten H-R (1999) Advances in analytical pyrolysis of soil
organic matter. J Anal Appl Pyrolysis 49:359-383

Lima DLD, Santos MS, Scherer WH, Schneider JR, Duarte CA, Santos BH,
Esteves IV (2009) Effects of organic and inorganic amendments on soil
organic matter properties. Geoderma 150:38-45

Mondini C, Cayuela ML, Sinicco T, Cordaro F, Roig A, Sánchez-Monedero MA
(2007) Greenhouse gas emissions and carbon sink capacity of amended
soils evaluated under laboratory conditions. Soil Biol Biochem 39:1366-
1374

Nardi S, Morari F, Berti A, Tosoni M, Giardini L (2004) Soil organic matter pro-
perties after 40 years of different use of organic and mineral fertilizers.
Eur J Agron 21:357–367

Negassa W, Baum C, Leinweber P (2011) Soil amendment with agro-industrial
products : molecular-chemical compositions and effects on soil bioche-
mical activities and phosphrous fractions. J Plant Nutr Soil Sci 174:113-
120

Nierop KGJ, Pulleman MM, Marinissen JCY (2001) Management- induced OM
differentiation in grassland arable soil : a study using pyrolysis tech-
niques. Soil Biol Biochem 33:755-764

Paul EA, Clark FE (1989) Soil microbiology and biochemistry.San Diego :
Academic Pr, 237 p

Piccolo A, Spaccini R, Nieder R, Richter J (2004) Sequestration of a biologically
labile organic carbon in soils by humified organic matter. Clim Change
67:329-343

Rivero C, Chirenje T, Ma LQ, Martinez G (2004) Influence of compost on soil
organic matter quality under tropical conditions. Geoderma 23:355-361

Sánchez-Monedero MA, Cayuela ML, Mondini C, Serramia N, Roig A (2008)
Potential of olive mill wastes for soil carbon sequestration. Waste Manag
28:767-773

Sánchez-Monedero MA, Serramia N, Civantos CGO, Fernandez-Hernandez A,
Roig A (2010) Greenhouse gas emissions during composting of two-
phase olive mill wastes with different agro-industrial by-products.
Chemosphere 81:18-25

Schulten H-R, Hempfling R (1992) Influence of agricultural soil management
on humus composition and dynamics : classical and modern analytical
techniques. Plant Soil 142:259-271

Schulten H-R, Leinweber P, Reuter G (1992) Initial formation of soil organic
matter from grass residues in a long-term experiment. Biol Fertil Soils
14:237-245

Smidt E, Eckhardt K-U, Lechner P, Schulten H-R, Leinweber P (2005) Charac-
terization of different decomposition stages of biowaste using FT-IR
spectroscopy and pyrolysis-fieldionization mass spectrometry. Biode-
gradation 16:67-79

Spaccini R, Sannino D, Piccolo A, Fagnano M (2009) Molecular changes in
organic matter of a compost amended soil. Eur J Soil Sci 60:287-296

Spaccini R, Piccolo A (2007) Molecular characterisation of compost at in-
creasing stages of maturity : II: Thermochemolysis-GC-MS and 13C-CP-
MAS-NMR spectroscopy. J Agr Food Chem 55:2303-2311

Stewart CE, Neff JC, Amatangelo KL, Vitousek PM (2011) Vegetation effects on
soil organic matter chemistry of aggregate fractions in a Hawaiian Forest.
Ecosystems 14:382-397

Stuczynski TI, McCarty GW, Reeves JB, Wright RJ (1997) Use of pyrolysis GC/MS
for assessing changes in soil organic matter quality. Soil Sci 162:97-105

Weber J, Karczewska A, Drozd J, Licznar M, Licznar S, Jamroz E, Kocowicz A
(2007) Agricultural and ecological aspects of a sandy soil as affected by
the application of municipal solid waste composts. Soil Biol Biochem
39:1294-1302

Zech W, Senesi N, Guggenberger G, Kaiser K, Lehmann J, Miano TM, Miltner A,
Schroth G (1997) Factors controlling humification and mineralization of
soil organic matter in the tropics. Geoderma 79:117-161

Zucconi F, Monaco A, Forte M (1985) Phytotoxins during the stabilization of
organic matter. In: Gasser JKR (ed) Composting of agricultural and other
wastes. London : Elsevier, pp 73-86