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Comparison of several maturity indicators for estimating phytotoxicity in compost-amended soil

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Abstract

Compost can provide a rich organic nutrient source and soil conditioner for agricultural and horticultural applications. Ideal compost amendment rates, however, vary based on starting material and compost maturity or their interaction, and there is little consensus on appropriate methods to gauge maturity. In this study, electrical conductivity, carbon-to-nitrogen ratio, and carbon mineralization measurements were made on compost-amended soils and compared to phytotoxicity measured as cress (*Lepidium sativum*) germination. Cress germination in soil and compost mixtures incubated for 8–10 days significantly decreased with increasing electrical conductivity and carbon mineralization rate of the mixture and with carbon mineralization rate and mineralizable carbon associated with the compost. Cress germination was not related to carbon-to-nitrogen ratio or pH of soil and compost mixtures. The electrical conductivity of the soil and compost mixtures significantly decreased with decreasing mineralizable carbon suggesting that compounds contributing to electrical conductivity were present in the compost and decomposed upon soil amendment. The results of this study indicate that measurements of mineralizable carbon and mineralization rate of composts in soil, and electrical conductivity and mineralization rate of soil and compost mixtures, can be used as indicators of compost maturity.

1. Introduction

Composting of the biodegradable portion of household, municipal, and agricultural wastes can reduce the volume of wastes going to landfill and provide a rich, organic nutrient source and soil conditioner for commercial agriculture (Goyal et al., 2005; Levy and Taylor, 2003; Smith and Hughes, 2001). Problems associated with compost phytotoxicity – the "intoxication of living plants by substances present or produced in the growth medium, when these substances are taken up and accumulated in plant tissue" (Araujo and Monteiro, 2005) – have impeded compost use in agriculture (Erhart and Burian, 1997; Inbar et al., 1990; Mondini et al., 2003; Morel and Guillemain, 2004). The source of materials and the biological activity associated with composting make it extremely difficult to assess the rate of compost application and its suitability as a soil amendment.

Maturity is a term used to indicate the level of phytotoxic substances in composts and compost suitability for plant growth (Benito et al., 2005; Brewer and Sullivan, 2003; Rynk, 2003). There is currently no universal method for measuring compost maturity (Goyal et al., 2005; Kato et al., 2005; Mondini et al., 2003; Provenzano et al., 2001; Rynk, 2003), in part due to the many causes of phytotoxicity. Some of the methods that have been used to measure maturity include carbon-to-nitrogen ratio (C/N), changes in nitrogen species, pH, electrical conductivity, cation exchange capacity, organic chemical constituents, reactive carbon, humification parameters, optical density, temperature, color, odor, structure, specific gravity, plant assays, respiration, microbial population changes, and enzyme activity (Epstein, 1997).

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Plant assays often are used for measuring compost maturity (Wu et al., 2000; Epstein, 1997; Chen and Inbar, 1993; Cooperband et al., 2003) and are especially beneficial because they can measure the compounded effects of various phytotoxic factors (Emino and Warman, 2004; Zucconi et al., 1981). However, plant assays vary greatly in their methodology (Emino and Warman, 2004; Warman, 1999), which has involved assessing compost extracts, direct seeding in compost, planting in a potting media containing compost, and using a variety of plant species in conjunction with employing various methods of evaluation (e.g., seed germination, seedling length, root length and biomass). These methods are not always consistently or sufficiently sensitive (Rynk, 2003; Emino and Warman, 2004; Warman, 1999) and can be time-consuming (Helfrich et al., 1998; Rynk, 2003; Cooperband et al., 2003). Recently, a consistent and sensitive plant assay involving the seeding of cress directly into compost and soil mixtures and measuring phytotoxicity via germination was developed (Aslam and VanderGheynst, 2008). Relating this assay of phytotoxicity to a readily measurable compost property could reduce the time needed to evaluate phytotoxicity and maturity.

Two additional methods often used for measuring compost maturity include C/N ratio and electrical conductivity (EC). Composts with high C/N ratio can cause nitrogen immobilization upon amendment to soil (Hoitink and Boehm, 1999) and those with low C/N ratio can cause ammonium toxicity (Epstein, 1997; Hoitink and Boehm, 1999). Researchers have suggested various ideal C/N ratios ranging from <12 to <25 (Brewer and Sullivan, 2003; Erhart and Burian, 1997; Jimenez and Garcia, 1992), but the optimal value is often dependent on the initial feedstock (Benito et al., 2005; Epstein, 1997). Electrical conductivity measures the concentration of soluble ions or the salinity of the compost. Excessive salinity in compost can cause phytotoxicity directly, depending on the salt tolerance of the plant species. Salinity also can develop from nitrogen mineralization and production of organic acids.

Respiration rate, measured by CO_2 evolution, represents carbon mineralization rate and is a fairly routine compost measurement. Respiration rate may be a good indicator of compost maturity because high respiration can lead to nitrogen immobilization, anaerobic conditions, and the formation of phytotoxic compounds (Benito et al., 2005; Chen and Inbar, 1993; Epstein, 1997; Jimenez and Garcia, 1992; Wu et al., 2000). Carbon mineralization rate of a compost and the amount of mineralizable carbon remaining upon amendment to soil also may be good indicators of phytotoxicity because many phytotoxic compounds decompose with time (Zucconi et al., 1981).

This study is part of a larger research effort to predict phytotoxicity in compost-amended soil from various maturity indicators (McEachin, 2006; Aslam and VanderGheynst, 2008). The focus of this paper is on which maturity indicators are of most value in this prediction. In this study, phytotoxicity is represented as cress seed germination and maturity indicators include electrical conductivity, C/N ratio, carbon mineralization rate, and potential mineralizable carbon in compost-amended soil. Results from food waste and green waste composts amended in sandy loam and potting soils are presented.

2. Materials and methods

2.1. Compost and soil preparation

Food waste and green waste composts were obtained from Jepson Prairie Organics (Dixon, California, USA) in December 2004. The food wastes were collected from San Francisco and Oakland, California's residential and commercial sectors including restaurants, delis, markets, coffee shops, hotels and bakeries. Food waste was composted under intermittent aeration in Ag-Bags (large plastic silage bags) for 30 days and then placed in windrows, where composts were turned and watered twice a week for an additional 30 days. Green waste was composted only in windrows for 60 days. Finished composts were screened through a 9.5-mm trommel screen prior to collection. Soils included a Reiff very fine sandy loam collected from the UC Davis student farm and a professional potting soil (Schultz Professional Potting Soil Plus, Spectrum Brands, Inc., St. Louis, Missouri, USA). The sandy loam was screened through a 3.18-mm sieve and stored dry at 4 °C.

Compost and soil moisture content were measured via methods published by the US Composting Council (Thompson, 2004). Electrical conductivity and pH were measured according to VanderGheynst and coworkers (VanderGheynst et al., 2004). Water holding capacity was measured by saturating the media, gravity draining for 24 h, and then estimating the moisture content of the saturated media by oven drying. Prior to the experiments, materials were wetted to 50% of their water holding capacities by gradual addition and mixing of distilled water (Table 1). The sandy loam was incubated in polyethylene bags at 50 °C for 24 h to kill weed seeds. Soils and composts were stored at –20 °C until needed to prevent microbial activity.

Table 1
Selected properties of composts, sandy loam and potting soils

Sample	Water holding capacity (% dry basis)	pH	EC (dS m^{-1})	C_T (mg CO_2–C g total $solids^{-1}$)
Food waste compost	208	5.5	6.65	390.3
Green waste compost	178	8.1	1.63	110.7
Sandy loam soil	44.5	6.8	0.089	1.26
Potting soil	391	5.9	0.283	6.79

2.2. Respiration measurement

Respiration measurements and compost carbon mineralization calculations followed those presented elsewhere (Aslam and VanderGheynst, 2008). Respiration measurements were made on 30-g dry weight samples of soil and soil/compost mixtures in 250-ml containers aerated continuously with humidified air at 20 ml/min to avoid oxygen limitations, and incubated at 35 °C (Thompson, 2004). Carbon dioxide concentration was measured using an infrared CO_2 sensor (Vaisala, Suffolk, UK). Data were recorded for each container every 5 h using a data acquisition system (VanderGheynst et al., 2002). Carbon dioxide evolution rate (CER) was calculated from a mass balance on each container. The total potential mineralizable carbon (C_T, Table 1) for each sample was determined by integration of the CER data (normalized by g total solids, gTS) with respect to time and regression using the following equation:

$$\int_0^t \overline{\mathrm{CER}}\,\mathrm{d}t = \frac{C_T * t}{c + t} \tag{1}$$

where t = time (days), CER (mg CO_2–C day^{-1} gTS compost^{-1}), C_T = total potential mineralizable carbon (mg CO_2–C gTS compost^{-1}), and c = constant (days).

Respiration measurements also were made on soils amended with compost at 0%, 5%, and 50% (v/v), where 0% provided a soil control and 5% and 50% represented field and horticulture applications, respectively. Six hundred milliliters of the mixtures was placed into heat-seal bags (Kapak, Fisher Scientific, Pittsburgh, Pennsylvania, USA), aerated continuously with humidified air at 20 ml/min and incubated at 35 °C. Carbon dioxide concentrations were measured on the influent and effluent air every 5 h using a data acquisition system (VanderGheynst et al., 2002), and CER was calculated from a mass balance on each bag. Three replicate bags were analyzed for each soil/compost mixture.

The CER data collected from aerated soil/compost mixtures over time, $\mathrm{CER_{Mixture}}$ (mg CO_2–C day^{-1} ml mixture^{-1}) and C_T estimates for each compost type, were used to calculate compost CO_2–C mineralization rate, $\mathrm{CER_{Compost}}$ (mg CO_2–C day^{-1} ml mixture^{-1}) and potential mineralizable compost CO_2–C remaining in the mixtures at any given time, C_R (mg CO_2–C ml mixture^{-1}). The CER data were divided by volume of media per bag since weights varied between treatments but volume was constant. The compost CO_2–C mineralization rate was estimated using the following equation:

$$\mathrm{CER_{Compost}}(t) = \mathrm{CER_{Mixture}}(t) - \mathrm{CER_{Soil}}(t) * V_{S/M} \tag{2}$$

where $\mathrm{CER_{Soil}}(t)$ = CO_2–C mineralization rate of the soil alone at time t (control) (mg CO_2–C day^{-1} ml mixture^{-1}), and $V_{S/M}$ = volumetric fraction of soil in the soil/compost mixture (ml soil ml mixture^{-1}).

The mineralizable compost CO_2–C remaining in the mixture was estimated by subtracting the cumulative compost CO_2–C evolved from the initial mineralizable compost CO_2–C amended given by

$$C_R(t) = C_T * R_C - \int_0^t (\mathrm{CER_{Compost}}(t))\mathrm{d}t \tag{3}$$

where R_C = initial concentration of amended compost in the mixture (gTS compost ml mixture^{-1}).

Incubation and media sampling times were selected according to respiration profiles of the compost-amended soil. The food waste compost mixtures were incubated for 10 days, and green waste compost mixtures were incubated for 8 days. Samples were taken from aerated bags initially, at a middle time point (4 days for food waste, and 3 days for green waste), and at the end of the experiments. Samples from replicate treatments were analyzed independently with the cress germination assay and EC measurements at each timepoint. The C/N was measured only on final samples. Samples from replicate treatments were pooled prior to C/N measurement. $\mathrm{CER_{Mixture}}$ and $\mathrm{CER_{Compost}}$ data were averaged for the first, middle (3rd or 4th day) and final days of incubation for comparison to the other measurements.

2.3. Carbon-to-nitrogen ratio measurements

Total carbon and nitrogen were determined by the University of California's Analytical Research Laboratory (DANR) using a dynamic flash combustion method coupled with a gas chromatographic separation system and a thermal conductivity detection system (Horwitz, 1997).

2.4. Plant assays

Plant assays were performed using a direct-seed test of garden cress ('Peppergrass', Lake Valley Seed Co., Boulder, Colorado, USA) in soil/compost mixtures (Aslam and VanderGheynst, 2008). Forty-five millimeters of mixture was added to Petri dishes, with 20 seeds per dish, and 2 assays were conducted per bag for each sampling time. Water controls consisted of 3 ml of distilled water in Petri dishes lined with #1 Whatman filter paper. Assays were initiated immediately upon bag sampling. Petri dishes were sealed with Parafilm to minimize water loss and placed in a wide- spectrum (GE Plant & Aquarium, Cleveland, Ohio, USA) lighted (71 microeinstein m^2 s^{-1}) incubator at 22 ± 2 °C with 10 h light/day (Warman, 1999) for 5 days. Seeds were examined for germination, where germination was defined as any protrusion through the seed coat. Percent normalized cress seed germination, $G_{N,\%}$, was calculated for each sample using the following equation:

$$G_{N,\%} = 100 * \frac{G_{\mathrm{Sample}}}{\overline{G}_{H_20}} \tag{4}$$

where G_{Sample} = number of cress seeds germinated in the assay and $\overline{G}_{H_20}$ = mean number of cress seeds germinated in the water control for the assay.

2.5. Experimental designs and statistical analysis

Completely randomized designs were used in all experiments. Numerical integrations of CER and $CER_{Compost}$ with respect to time and nonlinear regression of Eq. (1) were performed using KaleidaGraph v. 3.6 (Synergy Software, Reading, Pennsylvania, USA). Linear regressions were performed to determine if significant correlations existed between percent normalized germination and $CER_{Mixture}$, $CER_{Compost}$, C_R, EC and C/N ratio. Additional regressions were done to determine if EC was significantly correlated with $CER_{Mixture}$, $CER_{Compost}$, and C_R. Electrical conductivity, $CER_{Mixture}$, $CER_{Compost}$, and C_R were transformed using the natural logarithm to facilitate data symmetry. Analysis of variance (ANOVA) was used to determine the significance of the correlations. JMP-IN v. 5.1 (SAS Institute, Inc., Cary, North Carolina, USA) was used to perform linear regressions and ANOVA.

3. Results and discussion

Samples collected at the end of soil and soil/compost mixture incubations were used to investigate the relationship between normalized cress germination and EC, C/N ratio and carbon mineralization indicators (i.e., $CER_{Mixture}$, $CER_{Compost}$, and C_R). The EC of final samples increased with increasing compost amendment; EC varied between 0.23 and 2.95 dS m^{-1} for samples with potting soil and between 0.085 and 1.89 dS m^{-1} for samples with sandy loam (Table 2). The C/N ratio increased with increasing compost amendment for the sandy loam soil mixtures; however, the opposite was observed for the potting soil mixtures due to the very high C/N ratio of the potting soil relative to the compost. The carbon mineralization rates of the mixtures ($CER_{Mixture}$) also increased with increasing compost amendment and were above the soil controls indicating mixtures were still biologically active upon sampling. Normalized cress germination ranged between 0% for soils amended with 50% food waste compost and 100% for compost-free soils. The pH of mixtures after 8–10 days of incubation varied between 6.8 and 7.5 for sandy loam and 6.1 and 7.3 for potting soil (data not tabulated).

Correlations between percent normalized cress germination and C/N ratio, EC and carbon mineralization indicators were done for data listed in Table 2. Cress germination was not significantly correlated with C/N ratio ($P = 0.19$, $r = 0.41$), indicating this measurement is not appropriate for predicting phytotoxicity of the compost-amended soils investigated here. The lack of correlation was due in part to the potting soil having both a very high C/N ratio (ca. 60 g g^{-1}) and normalized germination in the absence of amended compost. The carbon in the potting soil was likely less labile compared to the carbon in the compost and contributed little to microbial activity and production of phytotoxic compounds. Similarly, Erhart and Burian (1997) did not find any correlation between C/N ratio and plant growth.

Despite the difference in EC among media (Table 1), normalized germination and the final EC of soils and soil/compost mixtures were significantly negatively correlated ($P = 0.0014$, $r = 0.81$). It has been reported in the literature that EC >2.45 dS m^{-1} is phytotoxic to cress (Sesay et al., 1997). Extracts from a 1/10 dilution of one soil/compost mixture exceeded and several others were within 1 dS m^{-1} of this EC. Soluble ions contributing to EC in extracts could have been from a variety of sources including inorganic salts associated with the soils and composts, volatile organic acids, ammonium and nitrate. Accumulation of nitrate is often a cause of phytotoxicity and it has been related to increases in EC (Smith and Doran, 1996). Although nitrate was not measured in this study, it could have been responsible in part for the increase in EC with

Table 2
Summary of measurements made on soils and soil/compost mixtures after 8 and 10 days of incubation at 35 °C

Soil[a]	Incubation time (days)	Compost level and type[b]	C/N (g g^{-1})	EC (dS m^{-1})	$CER_{Mixture}$[c] (mg CO_2–C day^{-1} ml^{-1})	$CER_{Compost}$[c] (mg CO_2–C day^{-1} ml^{-1})	C_R[c] (mg CO_2–C ml^{-1})	$G_{N,\%}$
PS	10	0%	56	0.28	0.08			82
PS	8	0%	64	0.27	0.12			103
PS	10	5% FW	48	0.73	0.25	0.17	3.4	71
PS	8	5% GW	51	0.43	0.22	0.11	0.8	86
PS	10	50% FW	20	2.81	2.00	1.96	39.3	12
PS	8	50% GW	22	1.65	1.15	1.09	7.3	33
SLS	10	0%	9	0.09	0.08			81
SLS	8	0%	9	0.09	0.09			89
SLS	10	5% FW	9	0.21	0.18	0.11	3.4	80
SLS	8	5% GW	12	0.12	0.21	0.13	0.9	67
SLS	10	50% FW	14	1.84	1.72	1.68	38.0	0
SLS	8	50% GW	14	0.57	1.05	1.01	8.1	54

Normalized germination ($G_{N,\%}$) was calculated from germination assays initiated within 5 h of sample collection.

[a] PS = potting soil and SLS = sandy loam soil.

[b] FW = food waste compost and GW = green waste compost.

[c] $CER_{Mixture}$ = mineralization rate of soil/compost mixture, $CER_{Compost}$ = mineralization rate associated with amended compost, and C_R = mineralizable carbon associated with amended compost.

compost amendment and mineralization. The pH of samples was well above the pK_a of acetic, butyric and proprionic acids (4.76–4.87) (Segel, 1976), which are volatile organic acids common in composts (Brinton, 1998). Therefore most volatile organic acids would have been in a dissociated form and could have contributed to EC.

Normalized germination was significantly correlated with carbon mineralization rate of the compost-amended soil mixtures ($CER_{Mixture}$) and carbon mineralization rate associated with compost ($CER_{Compost}$). Germination significantly decreased with increasing $CER_{Mixture}$ ($P < 0.0001$, $r = 0.89$) and $CER_{Compost}$ ($P = 0.0011$, $r = 0.92$). Normalized germination also was significantly negatively correlated with mineralizable carbon associated with the compost ($P = 0.0021$, $r = 0.91$). The decrease in germination could have been due to an increased production rate of phytotoxic compounds and the development of anaerobic environments. It could also be associated with the phytotoxic compounds present in the compost that decompose with time. Others also have reported an increase in phytotoxicity associated with high respiration

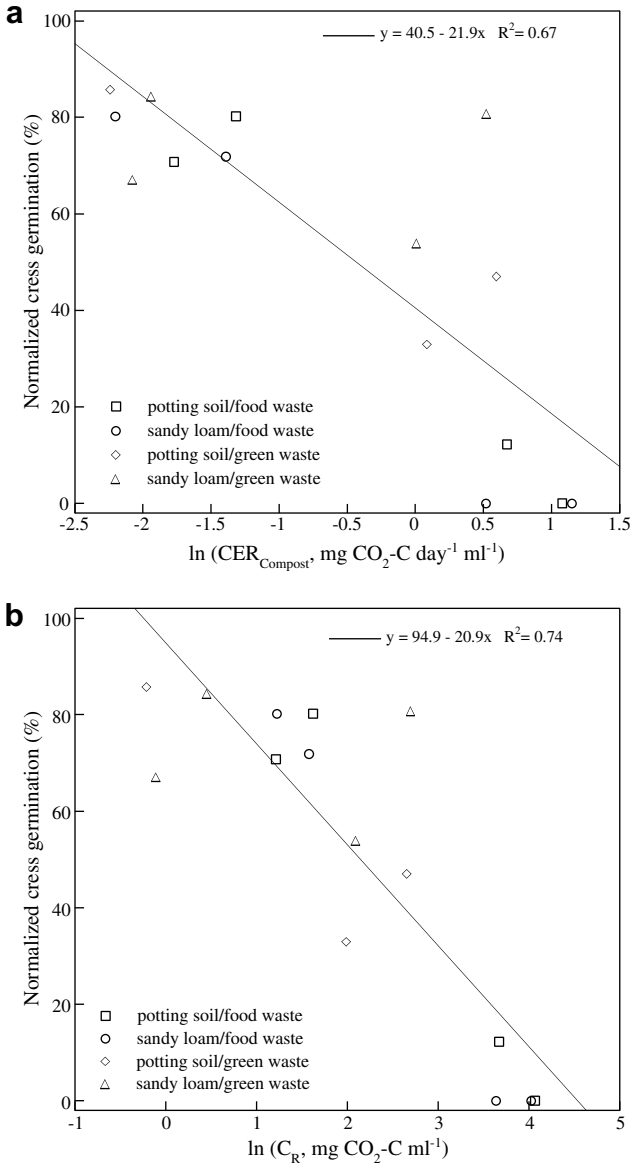

Fig. 1. Relationship between normalized cress germination ($G_{N,\%}$) for compost-amended soils (a) mineralization rate associated with amended compost ($CER_{Compost}$) and (b) mineralizable carbon associated with amended compost (C_R). Points represent data for samples taken from mixtures initially (C_R only), at a middle time point during incubation (3–4 days), and at the end of incubation (10 days for food waste and 8 days for green waste compost).

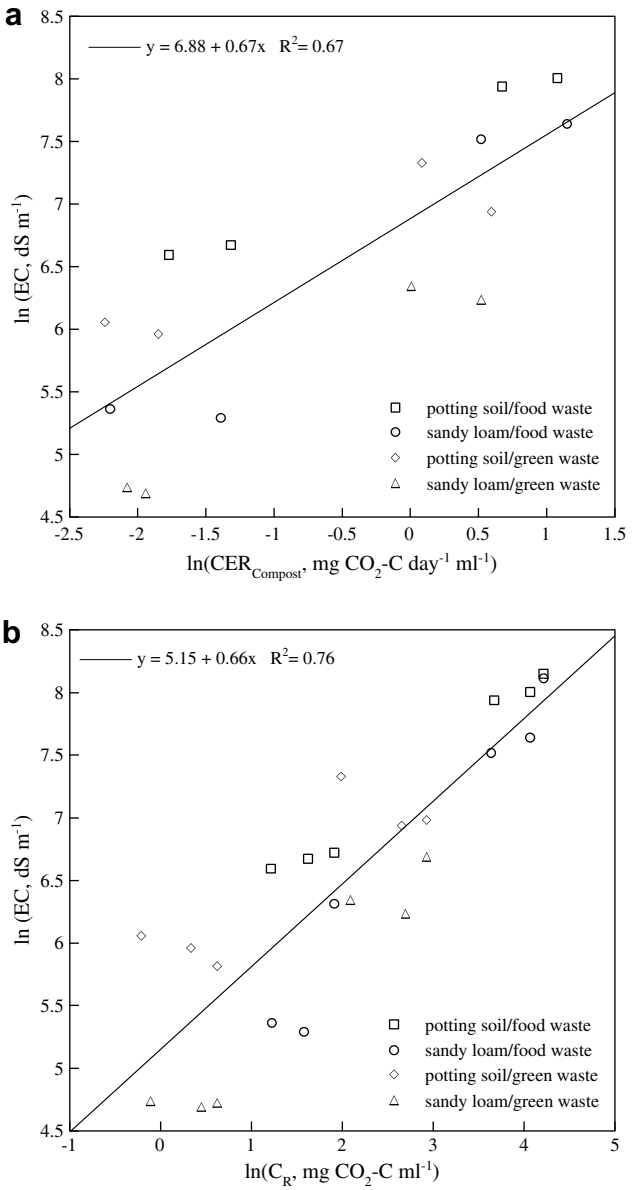

Fig. 2. Relationship between electrical conductivity (EC) for compost-amended soils (a) mineralization rate associated with amended compost ($CER_{Compost}$) and (b) mineralizable carbon associated with amended compost (C_R). Points represent data for samples taken from mixtures initially (C_R only), at a middle time point during incubation (3–4 days), and at the end of incubation (10 days for food waste and 8 days for green waste compost).

Table 3
Summary of pH measurements made on soils and soil/compost mixtures initially, at a middle time point during incubation (3–4 days), and at the end of incubation (10 days for food waste and 8 days for green waste compost)

Soil[a]	Amendment level (%)	Food waste compost		Green waste compost	
		Incubation time (d)	pH	Incubation time (d)	pH
PS	5	0	5.8	0	6.5
PS	5	4	6.4	3	6.6
PS	5	10	6.5	8	6.4
PS	50	0	5.5	0	7.3
PS	50	4	7.3	3	7.5
PS	50	10	7.1	8	7.3
SLS	5	0	6.5	0	7.1
SLS	5	4	6.7	3	6.9
SLS	5	10	6.9	8	7.0
SLS	50	0	5.5	0	7.9
SLS	50	4	7.0	3	7.3
SLS	50	10	7.2	8	7.5

[a] PS = potting soil and SLS = sandy loam soil.

activity of composts and have attributed phytotoxicity to similar factors (Benito et al., 2005; Chen and Inbar, 1993; Epstein, 1997; Jimenez and Garcia, 1992; Wu et al., 2000).

Since the correlations for $CER_{Compost}$ and C_R were based on very few samples ($n = 8$), additional data collected on initial (for C_R only) and middle time point soil/compost mixtures were used to confirm relationships (Fig. 1). Normalized germination was significantly negatively correlated with $CER_{Compost}$ and C_R ($P = 0.0003$, $r = 0.82$ and $P < 0.0001$, $r = 0.86$, respectively) confirming the negative correlation determined using final measurements from soil/compost mixtures. One data point for 50% green waste in sandy loam soil had a normalized germination much greater than other samples with similar respiration levels. One of the plant assay replicates in this treatment had unusually high germination. This illustrates that although the plant assay used is sensitive to phytotoxic compounds, it is still subject to variability and points to the need for a less variable maturity indicator.

EC was also regressed against $CER_{Compost}$ and C_R (Fig. 2) and $CER_{Mixture}$ (data not shown) to determine if the ions responsible for elevating EC were associated with increasing microbial activity (increasing $CER_{Mixture}$ and $CER_{Compost}$) and if EC decreased with increasing stabilization time (decreasing C_R). EC was significantly correlated with $CER_{Mixture}$ and $CER_{Compost}$ ($P = 0.0001$, $r = 0.81$ and $P = 0.0001$, $r = 0.82$, respectively) and highly significantly correlated with C_R ($P < 0.0001$, $r = 0.87$). Since the soils were not leached during incubation, the results suggest that the initial elevated EC was due to ions associated with compounds present in the composts that decomposed or volatilized during incubation of the mixtures. Volatile organic acids are common in immature food waste composts (Brinton, 1998) and the low pH of the food waste compost investigated here (i.e., 5.5) suggests the presence of organic acids. If volatile organic acids volatilized or decomposed during incubation, the pH of the mixture should increase with time. The pH did tend to increase with incubation time for mixtures containing food waste compost; however, no consistent change in pH was observed for soils amended with green waste compost (Table 3).

The significant relationships between germination and the other maturity indicators suggest that phytotoxicity can be predicted from measurements other than plant assays. While EC, $CER_{Mixture}$, $CER_{Compost}$ and C_R were all related to germination, they did require several procedures to measure. EC and $CER_{Mixture}$ also required measurements be made in soil and would be difficult to predict as compost stabilizes in soil. An advantage of C_R over the other measurements is that C_R in soils can be predicted using first-order kinetics (McEachin, 2006). The reported first-order model is based on potential mineralizable carbon in compost (e.g., C_T measured using respirometry prior to soil amendment), compost amendment rate, and soil temperature, and is independent of soil type. Employing such a model would allow producers to tailor compost production to achieve a certain C_T, or end users to adjust amendment rates and stabilization times upon soil amendment to minimize phytotoxicity.

4. Conclusions

Cress seed germination in soil and compost-amended soil mixtures decreased as EC, carbon mineralization rate associated with the soil and compost mixture ($CER_{Mixture}$), carbon mineralization rate associated with amended compost ($CER_{Compost}$), and mineralizable carbon associated with amended compost (C_R) increased. The correlation of cress germination with these four measurements was significant, suggesting these measurements could be used to determine phytotoxicity in compost-amended soil. The C/N ratio of soil/compost mixtures was not correlated with germination, which may have been due to the high background C/N ratio of the soils. While the change in pH was small in green waste compost, pH dynamics in soils amended with food waste compost could provide insight into possible mechanisms of phytotoxicity.

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