

ELIMINATION OF PHYTOTOXICITY DURING CO-COMPOSTING OF SPENT PIG-MANURE SAWDUST LITTER AND PIG SLUDGE

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Abstract

A plant seed germination technique was used to assess the phytotoxicity of spent pig litter–sludge at different stages of composting in forced-aerated piles on the relative seed germination, relative root elongation, and germination index (GI, a factor of relative seed germination and relative root elongation) of Chinese cabbage (*Brassica parachinensis*) and Chinese spinach (*Amaranthus espinosus*). The relative seed germination and root elongation of the two plants were significantly retarded by the spent litter–sludge extracts at day 0, but their values increased as composting progressed. From day 49 onwards, their values were similar to that of the control (deionized water) (between 80 and 100%). Increases in both relative seed germination and root elongation of the two plants corresponded with decreases in the concentrations of $\text{NH}_4^+\text{-N}$, water-extractable Cu and Zn of the compost demonstrating that these chemical compounds were gradually eliminated during composting. The multiple regression analyses showed that the $\text{NH}_4^+\text{-N}$ and water-extractable Cu concentrations in the spent litter–sludge extracts were the most important chemical factors causing the phytotoxicity. Composting at the top of the forced-aerated pile was slower than the middle, bottom and surface of the pile during the first 49 days of composting. However, from day 49 onwards, there was no difference among the four locations of the spent litter–sludge pile in terms of both plant responses and chemical parameters. These suggest that the spatial variations in the forced-aerated piles, in terms of phytotoxicity, gradually disappeared as the spent litter–sludge became mature. © 1998 Elsevier Science Ltd. All rights reserved

Key words: Forced-aerated composting, pig waste, plant bioassay, heavy metals, Ammonium.

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INTRODUCTION

Most studies show that animal manure compost exercises a positive influence on crop production due to its ability to supply nutrients to plants, and it also improves the physical properties of the soil (McConnell *et al.*, 1993; Tam & Wong, 1995; Chen *et al.*, 1996). Nevertheless, negative effects, normally associated with a decrease in yield and inhibition of seed germination and plant growth, have also been reported due to application of immature composts (Terman *et al.*, 1973; Tiquia *et al.*, 1996). Without stabilisation, the decomposition in the immature composts continues, foul odours develop, and metabolites are produced which are toxic to plants (Zuconi *et al.*, 1985). The harmful effects caused by the application of immature composts can, however, be avoided by composting the animal waste until the organic matter has become stabilised.

In the present study, the spent litter (a mixture of partially decomposed pig manure and sawdust) was co-composted with the pig sludge that settled at the bottom of the sedimentation tanks in the waste treatment plant. These two wastes were co-composted in the attempt to increase the nutrient content in the spent litter, and supply an adequate amount of organic matter to the pig sludge, to improve the beneficial value of these two wastes. Our previous work on composting of spent litter revealed that the water-extractable heavy metals decreased significantly with increasing humic substances as the spent litter reached maturity (Tiquia *et al.*, 1996, 1997a, 1997b). It was, therefore, worthwhile to explore the possibility of reducing the phytotoxic substances in the spent litter and pig sludge, such as heavy metals (Cu and Zn, in particular), by co-composting. To reduce the labour cost and space requirement for conventional windrow composting a forced-aerated composting method has been developed and examined. Since

forced-aerated composting is a non-turning method, the composting efficiency at different locations in the pile may be different due to variations in aeration level. Therefore, the phytotoxicity at different locations in the compost pile (top, middle, bottom and surface) was also determined. Seed germination and root elongation tests were employed to examine the phytotoxicity of the spent litter–sludge during composting.

METHODS

The spent litter (a mixture of partially decomposed sawdust and pig manure) disposed from the pig-on-litter system was mixed homogeneously with pig sludge at a ratio of 2:1 (spent litter:sludge; wet volume). The pig sludge was collected from the primary sedimentation tank of the waste treatment plant at Ta Kwu Ling Pig Breeding Centre, New Territories of Hong Kong. The moisture content of the mixture (spent litter–sludge) was 65% (w/v) at the beginning of composting. Three spent litter–sludge piles were built on perforated pipes connected to an air pump. Each pile was triangular in shape, about 2 m in width at the base and 1.5 m in height. Polyvinyl chloride (PVC) pipes (20 mm diameter) were laid at the base of the pile, with perforations (25 mm diameter) facing upward. The distance between each perforation was 20 cm. The pipes were covered by wood chips to prevent blockage of the holes, and air to the piles (from bottom to the top) was supplied by a Cole Palmer air pump, with an average flow-rate of 634 l min⁻¹ and a maximum output of 764 l min⁻¹. The air pump was on continuously during the entire period of composting. The spent litter–sludge piles were then topped off with a 5 cm layer of mature spent litter-compost to insulate the piles, and act as a biofilter to minimise odours. Spent litter–sludge samples were taken at four different locations in the pile: top (130 cm from the base of the pile), middle (75 cm from the base of the pile), bottom (30 cm from the base of the pile), and surface (5 cm from the surface of the pile) at day 0, and then weekly until the end of the composting process (day 77).

The spent litter–sludge samples were analysed for concentrations of total and water-extractable (1:10 w/v sample:water extract) Cu and Zn by atomic absorption spectrophotometry; NH₄⁺-N and (NO₃⁻+NO₂⁻)-N (using a distillation method; Sparks, 1996); water-extractable C (by TOC analyser, Shimadzu TOC-500); and electrical conductivity (EC; Sparks, 1996). The concentrations of these chemical properties were expressed on a 105°C dry weight basis. Aqueous extracts of the spent litter–sludge were prepared by shaking 10 g subsamples with 100 ml distilled water in a medical flat bottle for 1 h, using a horizontal shaker, then filtered. The phytotoxicity of these extracts was

evaluated by the seed germination technique (Tam & Tiquia, 1994; Tiquia *et al.*, 1996). Two plant species, namely Chinese cabbage (*Brassica parachinensis*), and Chinese spinach (*Amaranthus spinosus*) were used for the test. After 5 days of incubation in the dark, the seed germination percentage and root length of the two plant species in the extracts were determined. The seed germination percentage and root elongation of the two plant species in deionized water were also measured and used as the control. A 5 mm primary root was used as the operational definition of seed germination (USEPA, 1982). The percentages of relative seed germination, relative root elongation and germination index (GI) were calculated as follows:

Relative seed germination(%)

$$= \frac{\text{No. of seeds germinated in litter extract}}{\text{No. of seeds germinated in control}} \times 100$$

Relative root growth (%)

$$= \frac{\text{Mean root length in litter extract}}{\text{Mean root length in control}} \times 100$$

$$\text{GI} = \frac{(\% \text{ Seed germination}) \times (\% \text{ Root growth})}{100}$$

The cause and effect of the chemical properties of the spent litter–sludge on the GI values of the two plant species were evaluated by regression analyses. Both linear and curvilinear regression models were used. A stepwise multiple regression analysis was also performed to determine the most important chemical factor affecting seed germination, root elongation and GI. All statistical analyses executed in the present study were based on the procedures suggested by Zar (1984).

RESULTS AND DISCUSSION

Phytotoxicity assay and chemical properties of the spent litter–sludge

The responses of the two plants to the toxicity of the spent litter–sludge extracts in terms of the relative seed germination percentages differed at day 0 [Fig. 1(a) and (b)], but the relative seed germinations of both species increased to about 100% and 90% by day 77. The responses to the toxicity in terms of the relative root elongations were similar [Fig. 1(c) and (d)], showing a temporary retardation at the beginning of composting. The root elongation then increased to between 80 and 100% by the end of composting. In the spent litter–sludge collected at the top location of the pile, the increases in both seed germination and root elongation of the two plants were slower than those in the middle, bottom

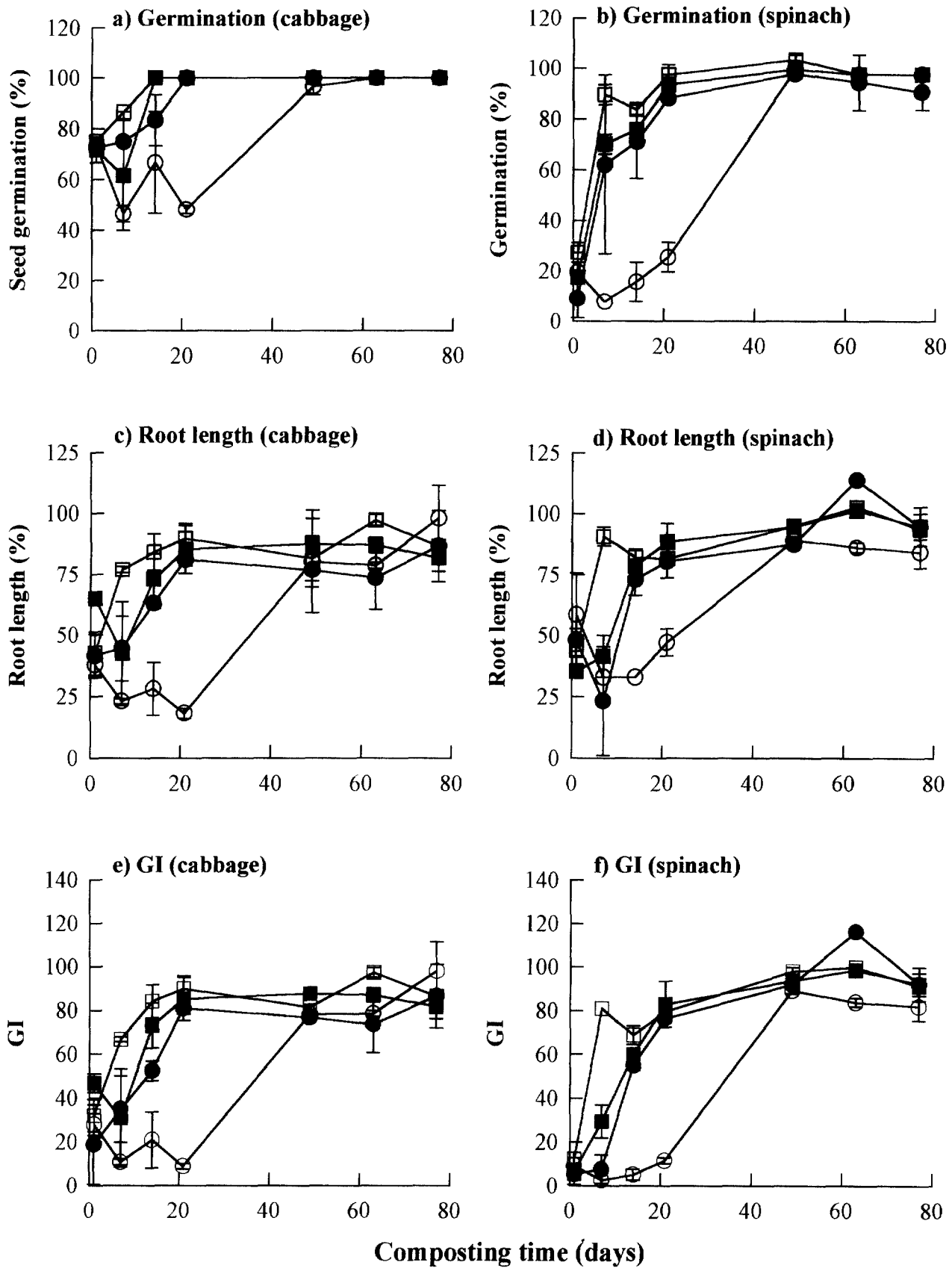


Fig. 1. Relative seed germination, relative root elongation and germination index (GI) of Chinese cabbage and Chinese spinach in the spent litter–sludge extracts at different stages of composting. (○ = top; ● = middle; □ = bottom; ■ = surface locations of the compost pile; mean and standard deviation of three replicates are shown.)

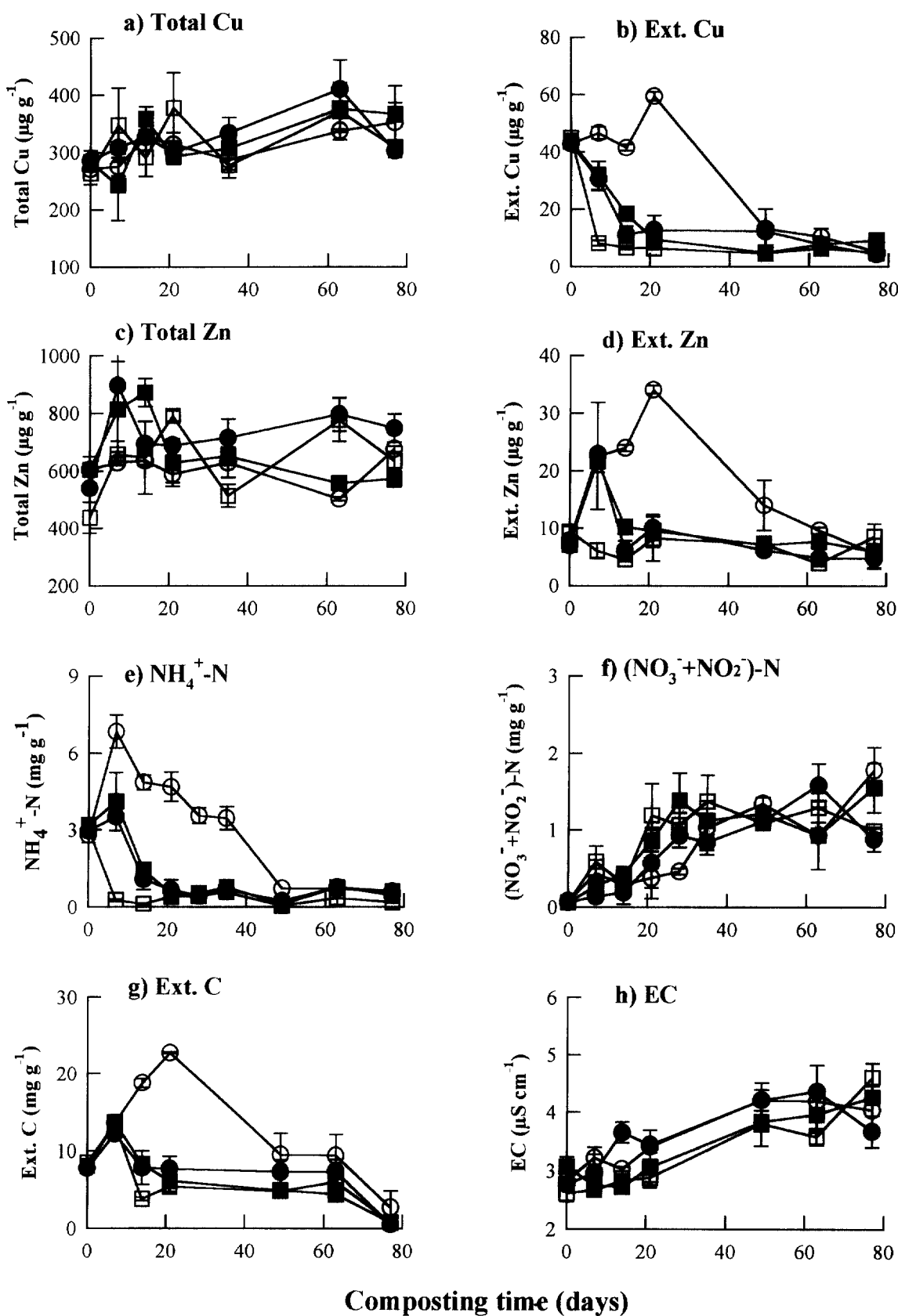


Fig. 2. Changes in chemical properties of the spent litter-sludge during the composting process. (○ = top; ● = middle; □ = bottom; ■ = surface locations of the compost pile; mean and standard deviation of three replicates are shown.)

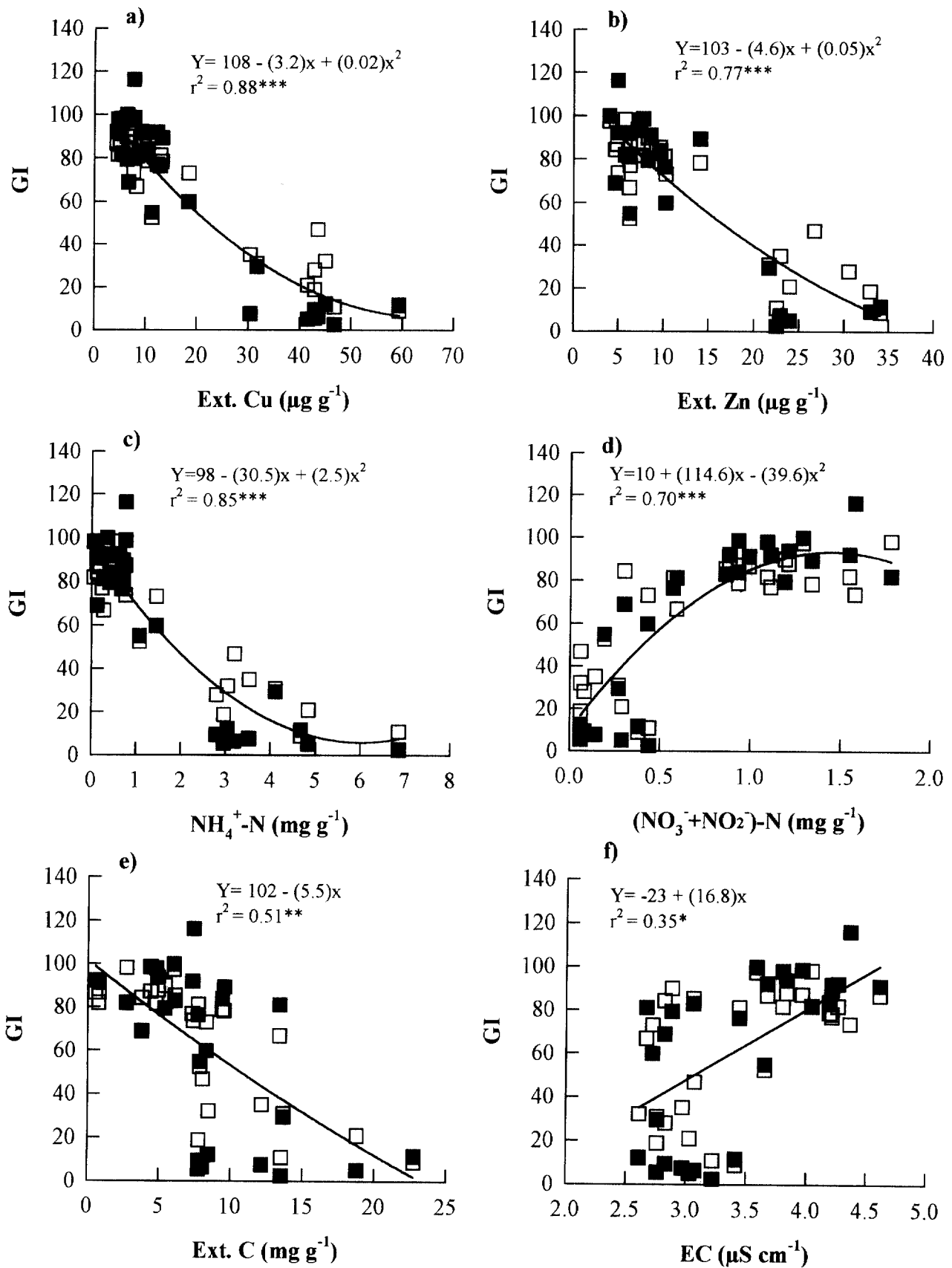


Fig. 3. Relationships between germination index (GI) and chemical properties of the spent litter-sludge. (\square = Chinese cabbage; \blacksquare = Chinese spinach.)

Table 1. Multiple regression analyses of phytotoxicity assays and five chemical properties of the spent litter extract

Plant species	Multiple regression equation	Multiple R^2 value	Adjusted R^2 value	F value	Significance of F
Seed germination					
Cabbage	%GERM = $104.2 - (6.2 * \text{NH}_4) - (0.3 * \text{Ext. Cu})$	0.92	0.91	140.9	<0.0001
Spinach	%GERM = $94.2 - (4.4 * \text{NH}_4) - (0.73 * \text{Ext. Cu}) - (0.4 * \text{Ext. Zn})$	0.91	0.89	96.6	<0.0001
Root elongation					
Cabbage	%ROOT = $96.9 - (1.1 * \text{Ext. Cu}) - (0.7 * \text{NH}_4)$	0.89	0.88	102.0	<0.0001
Spinach	%ROOT = $94.7 - (12.5 * \text{NH}_4)$	0.79	0.78	96.7	<0.0001
Germination index					
Cabbage	%GI = $92.94 - (5.8 * \text{NH}_4) - (1.0 * \text{Ext. Cu})$	0.91	0.90	122.5	<0.0001
Spinach	%GI = $92.04 - (18.6 * \text{NH}_4)$	0.78	0.77	93.95	<0.0001

Regression equation was calculated based on five chemical parameters (water-extractable Cu and Zn, $\text{NH}_4^+\text{-N}$, and $(\text{NO}_3^- + \text{NO}_2^-)\text{-N}$ and EC), using STEPWISE METHOD and PIN (Probability to F-enter) = 0.050 limit; NS: not significant at PIN = 0.05 limit).

GERM, germination; ROOT, root elongation; GI, germination index; Ext., extractable.

and surface parts during the first 49 days of composting, indicating that the elimination of phytotoxicity in this top location was slow. From day 49 onwards, the seed germination and root elongation values among the four locations of the spent litter-sludge pile were similar. Their values were also similar to that of the control (deionized water).

It has been suggested that a germination index (a factor of relative seed germination and relative root elongation) of $\geq 80\%$ indicated the disappearance of phytotoxins in composts (Zucconi *et al.*, 1981; Tiquia *et al.*, 1996). In the present experiment, such a value was reached by day 49 for both plants [Fig. 1(e) and (f)]. Increases in the GI values of the test plants corresponded with the decreases in concentrations of $\text{NH}_4^+\text{-N}$, water-extractable Cu and Zn (Fig. 2b, d, and e), suggesting that $\text{NH}_4^+\text{-N}$ and heavy metals in the spent litter-sludge were major compounds inhibiting seed germination and root growth. The decrease in $\text{NH}_4^+\text{-N}$ concentration in the spent litter-sludge to low levels was associated with the accumulation of $\text{NO}_3^-\text{-N}$ via the nitrification process (Fig. 2e and f). On the other hand, the drop in water-extractable Cu and Zn concentrations could be attributed to the formation of a metal-humus complex, thus making these two metals not water-extractable and biologically unavailable. The phytotoxicity of composts can also be due to high electrical conductivity (EC) content ($> 4 \mu\text{S cm}^{-1}$) (Allison, 1973). Nevertheless, the high EC content recorded at the end of composting ($3.5\text{--}4.7 \mu\text{S cm}^{-1}$) did not cause any retardation of seed germination and root elongation of the plants examined [Figs 1(a), (b) and 2(h)]. In fact, increases in EC during composting corresponded with increases in GI values of the plants in this present study [Fig. 3(f)]. Such a finding implies that both species have a wide tolerance to high EC content in the spent litter-sludge extracts.

The decreases in concentrations of $\text{NH}_4^+\text{-N}$, water-extractable Cu and Zn in the samples collected at the top location of the pile were the slowest. This is

probably due to the fact that the aeration level in the top portion of the pile was less than at the other three locations, so the decomposition was slow. However, after day 49, these values were similar to the samples collected in the other three locations [Fig. 2(b), (d) and (e)] suggesting that the spatial variations in the forced-aerated piles disappeared as the spent litter-sludge became mature.

Relationship between germination index (GI) and the chemical properties of the spent litter-sludge extracts

The results of the present study fitted the curvilinear regression models better than the simple linear regression. Curvilinear relationships between GI and the water-extractable chemical parameters are shown in Fig. 3. The magnitude of changes in GI values was strongly dependent on the chemical properties of the spent litter-sludge (Fig. 3). A GI value of around 80–100% was achieved when the chemical concentrations were: $\leq 15 \mu\text{g g}^{-1}$ water-extractable Cu, $\leq 20 \mu\text{g g}^{-1}$ water-extractable Zn, $\leq 1 \text{ mg g}^{-1}$ $\text{NH}_4^+\text{-N}$ and $\leq 10 \text{ mg g}^{-1}$ water-extractable C (Fig. 3). Increases in $(\text{NO}_3^- + \text{NO}_2^-)\text{-N}$ content of the spent litter-sludge to around $1.0\text{--}2.0 \text{ mg g}^{-1}$ also exerted a positive influence on the GI [Fig. 3(d)]. When these values are reached during composting, the phytotoxicity in the spent litter-sludge in terms of these chemicals is eliminated.

The multiple regression analyses showed that the $\text{NH}_4^+\text{-N}$ concentration of the spent litter-sludge was the most important chemical factor affecting germination of the plants tested. Ammonium has always been found to be the most important factor in the phytotoxicity of spent pig litter (Tiquia *et al.*, 1996). In the present study, the $\text{NH}_4^+\text{-N}$ concentration was also found to be the most important chemical component influencing root elongation of Chinese spinach. However, the decrease in water-extractable Cu concentration was more related to the increase in root elongation of Chinese cabbage than was

$\text{NH}_4^+\text{-N}$ concentration (Table 1), thus the Cu^{2+} content was considered as the most important factor affecting root elongation of this species. This result is inconsistent with that obtained from our previous study on the spent litter, which reported that $\text{NH}_4^+\text{-N}$ was the most important factor inhibiting root elongation of all species examined including Chinese cabbage (Tiquia *et al.*, 1996). Such a discrepancy could be due to the different initial $\text{NH}_4^+\text{-N}$ levels in the spent litter–sludge (3 mg g^{-1}) and in the spent pig litter ($4\text{--}8 \text{ mg g}^{-1}$) reported in our previous work (Tiquia *et al.*, 1996; Tiquia *et al.*, 1997a).

Other compounds such as acetic, propionic, butyric and isobutyric acids, may be possible inhibitors of seed germination and root elongation (Zuconi *et al.*, 1985; Epstein, 1997). The phytotoxic character of these chemical compounds has not yet been investigated on the spent litter–sludge and further work will be concentrated on this aspect.

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