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Tomato growth in soil amended with sugar mill by-products compost

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Abstract

Sugar mill by-products compost may be a good soil amendment to promote tomato (*Lycopersicon esculentum* L.) growth. In addition, the compost may further promote plant growth by inoculation with N_2-fixing bacteria. Compost from sugar-mill waste was prepared with and without the N_2-fixing bacteria, *Azotobacter vinelandii*, *Beijerinckia derxii* and *Azospirillum* sp. and incubated for 50 days. Each compost type was added to 10 kg of soil in pots at rates of 0, 15, and 45 g with and without fertilizer N at rates of 0, 0.75, and 1.54 g. A blanket application of P and K was applied to all pots. Shoot and root dry weights and N content of the whole plant was measured at 55 days. Dry weight of tomato shoots was increased by 40% by addition of fertilizer N and root weight was increased by 66%. Without fertilizer N the high rate of inoculated compost increased shoot growth 180% and uninoculated compost increased shoot growth 112%. For most treatments with and without fertilizer N, inoculated compost enhanced shoot growth and nitrogen content more than uninoculated compost. Root weights were nearly doubled by addition of either compost in comparison to the 0 N treatment. At the low rate of compost addition without fertilizer N, root weight was the same for uninoculated and inoculated compost but at the high rate of compost addition root weight was 32% higher for inoculated compost. The N_2-fixing bacteria colonized roots when inoculated compost was used. Sugar mill by-products compost proved to be an effective soil amendment for promoting the growth of tomato plants.

Introduction

Thailand has 46 sugar mills producing approximately 20 million tonnes of filter cake and bagasse annually as waste products that may be composted into a soil amendment providing a good source of N, P and Ca (Meunchang et al., 2005a, b). Filter cake, when composted with bagasse, helped conserve N and it seemed to be suitable for agronomic use because of its neutral pH, low phyto-toxicity and nutrient content (Meunchang et al., 2005a). Inoculation of the compost with the plant growth promoting rhizobacteria, *Beijerinckia, Azotobacter*, and *Azospirillum*, increased the available P content by approximately 25% and the N gain by up to 16% (Meunchang et al., 2005b). The rhizobacteria showed good survival in the compost with populations in the range of 1×10^4 g^{-1} of compost (Meunchang et al., 2005b).

Direct inoculation of soil or plants with N_2-fixing bacteria or plant growth promoting rhizobacteria has been extensively studied and reviewed by Vessey (2003). He surmises that for nonlegumes there is some evidence that enhanced plant growth may be achieved due to N_2-fixation but it is most likely from production of plant growth stimulating substances. Inoculated and uninoculated sugar mill by-products compost has

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not been evaluated to determine their impact on plant growth.

Tomato is a high value crop in Thailand, both in the field and under glass house conditions (Opeña, 1985) with annual production of approximately 250,000 tonnes (Department of Agricultural Extension, 2005). Soil amendments with the sugar mill by-product compost may have advantages for tomato production as well as environment viewpoints.

The primary objective of our investigation was to determine the ability of sugar mill by-products compost and sugar mill compost inoculated with N_2-fixing bacteria to promote growth of tomatoes grown in pots. A secondary objective was to determine the ability of the rhizosphere bacteria in compost to colonize tomato roots.

Materials and methods

Experimental approach

A glasshouse experiment was designed (three replications) to determine the impact of inoculated and uninoculated compost on tomato growth with and without added fertilizer N. To help to determine the N sufficiency of the soil for tomato growth three rates (0, 0.75 and 1.5 g N pot^{-1} that was applied in two split applications of half at each application) of fertilizer N were included as control treatments without compost. The experimental units were 45 (30 cm diameter) clay pots filled with 10 kg of soil. The soil was a Nam Pong loamy sand (Thai soil series) with characteristics as provided in Table 1. Two compost types, inoculated and uninoculated, were used at rates of 15 and 45 g dry weight per pot. Control treatments receiving the three N rates but no compost were included for help in determining the impact of composts on providing N.

Compost

Compost was prepared by mixing filter cake and bagasse (2:1 by weight) collected from Mitr Kasetr Industry Co., Ltd. Tanaka, Kanchana Buri province, Thailand. The composting process was operated for 40 days at which time loss of organic matter stabilized (Meunchang et al., 2005a). The compost was separated into two parts, the one part was inoculated with N_2-fixing bacteria and leaving the second part uninoculated. The characteristics of uninoculated and inoculated compost are provided in Table 1.

Table 1. Physico-chemical and biological characteristics of uninoculated (CC) and inoculated (CN) sugar mill by-products compost after 50 days of incubation and a Nam Pong soil

Parameter	CC	CN	Soil
Physico-chemical			
pH (compost; water 1:5, soil; water 1:1)	7.9	7.3	6.0
Total organic matter (g kg^{-1})	356	385	4.8
Total Nitrogen (g kg^{-1})	18.6	20.5	0.7
Total Phosphorus (g kg^{-1})	13.5	12.3	0.02
Available P Bray II (mg kg^{-1})	257	304	5.0
Total Potassium (g kg^{-1})	5.2	5.4	0.05
Exchangeable Potassium (mg kg^{-1})	2450	3280	3.5
Total Calcium (g kg^{-1})	100.0	99.0	0.26
Exchangeable Calcium (mg kg^{-1})	232	464	12.8
Biological properties (cfu g^{-1} fresh weight)			
Azotobacter vinelandii ATTC 478	< 10	2.4×10^3	< 10
Beijerinckia derxii ATCC 49360	< 10	2.1×10^4	< 10
Axospirillum sp. TS8	< 10	1.8×10^5	< 10

The N_2-fixing bacteria were: *Azosporillum* TS8 (Meunchang et al., 2004), *Azotobacter vinelandii* ATCC 478 and *Beijerinckia derxii* ATCC 4936. Composting was continued for 50 days after inoculation. Numbers of inoculated bacteria in compost after incubation for 50 days were estimated. *Azotobacter* and *Beijerinckia* were determined with the dilution plate method on pH and carbon specific N-free medium (Dobereiner, 1980) and *Azospirillum* was determined with dilution and enumeration from its pellicle formation approximately 0.3 mm under the surface of N-free semi-solid medium (NFb) (Dobereiner, 1980).

Planting and fertilization

Tomato (*Lycopersicon esculentum* L. variety Valentile 183 hybrid, CP Co., Ltd.) seedlings were grown for 20 days in an unfertilized 1:1 volume mixture of composted rice hulls and sterilized washed fine sand. One seedling was transplanted into each clay pot containing soil. Compost had already been thoroughly mixed at pre-determined rates into the soil before it was filled into the pots. N, P and K chemical fertilizers based on a recommendation of the Thailand Department of Agriculture were applied into the potting mixture 5 cm away from the plant stem and mixed well within the top 5 cm of soil 10 and 20 days after transplanting. Phosphorus (0.7 g P pot^{-1}) and K (1.0 g K pot^{-1}) were applied at the time of each fertilizer N application. The N fertilizer was urea, the P fertilizer was triple super phosphate and the K fertilizer was potassium chloride. Plants were irrigated with tap water that had been passed through a mixed cation/anion exchange resin, boiled and cooled.

Plant analyses

Plants were harvested 55 days after transplanting. Shoots with eventual green fruit and roots were thoroughly washed, dried at 65 °C for 72 h and weight was recorded. Shoots with green fruit and roots were combined and ground to approximately 0.5 mm for N, P, Ca, and K analyses. Nitrogen was determined by the micro-Kjeldahl method following digestion of plant material in sulfuric acid with catalysts (Page et al., 1982).

Nitrogen-fixing bacteria on roots

N_2-fixing bacterial numbers on the roots were counted at 55 days by placing 5 g of fresh roots into 45 ml of sterile nitrogen free NFb mineral solution medium. *Azotobacter* and *Beijerinckia* were measured by making serial dilutions and spread plating on N-free LG and N-free NB media (Döbereiner, 1980), respectively. *Azospirillum* was measured by most probable number (MPN) techniques according to Zuberer (1994) in a selective N_2-free NFb semi-solid medium (Döbereiner, 1980).

Statistical analyses

The compost treatments were analyzed as a factorial design without the control treatments not receiving compost so that interactions between compost type, compost rates, and fertilizer rates could be determined. Additional statistical analyses were made including the control treatments without compost. This was accomplished by considering the control treatment as a compost type in a factorial arrangement of treatments for each compost rate. The statistical software package used for the various analyses was Statistix7 from Analytical Software of Tallahassee, Florida, USA.

Results

The treatments of N fertilizer addition, compost type, and compost rate all significantly influenced shoot weight (Table 2). There were statistically significant two-way and three-way interactions between the main factors of compost type, compost rate, and nitrogen rate on shoot weight (Table 2). This indicates that each of the main factors did influence shoot mass but to understand their impacts the individual treatment combinations must be examined. Table 3 provides data for comparing compost treatments with each other and with the control treatment not receiving compost. The data for the control treatment, no compost addition, are the same in Table 3, for both rates of compost addition because the same controls were used for both rates. For treatments receiving compost, the higher rate of compost addition consistently

Table 2. Summary of probabilities for significant differences between compost type, compost rate, and nitrogen rate on shoot and root weights and N percentage in the whole plant

Source of variation[a]	df	Shoot weight	Root weight	N%
Compost (A)	1	0.00	0.06	0.00
Compost rate (B)	1	0.00	0.00	0.00
Nitrogen (C)	2	0.00	0.45	0.00
A×B	1	0.00	0.28	0.44
A×C	2	0.00	0.64	0.09
B×C	2	0.00	0.87	0.28
A×B×C	2	0.06	0.44	0.76

[a]The treatment not receiving compost was not included in this analysis.

Table 3. Shoot weight of tomato as influenced by N rate, compost type, and compost rate

Nitrogen rate	Compost	Compost rate/pot[a]	
		15 g g/plant	45 g
0	None[b]	58.1f	58.1 f
0.75	None	66.5 e	66.5 e
1.54	None	81.7 d	81.7 d
0	Uninoculated	61.6 of	108.7 c
0.75	Uninoculated	65.6 e	119.4 b
1.54	Uninoculated	91.2 c	131.6 a
0	Inoculated	84.3 cd	123.7 b
0.75	Inoculated	105.8 b	134.6 a
1.54	Inoculated	115.4 a	135.3 a

[a]Numbers within columns not having letters in common were significantly different at P (0.05).
[b]The treatment not receiving compost was included in the analysis of variance for comparative purposes.

increased shoot weight over the lower rate for the same fertilizer N addition (Table 3).

The highest rate of N addition increased shoot mass 41% for the treatment not receiving compost. Fertilizer N addition also increased shoot weight for both compost types at both rates of compost addition. Uninoculated compost added at the higher application rate without fertilizer N produced 33% more shoot weight than the highest rate of fertilizer N without compost. Inoculated compost added at the higher application rate without fertilizer N produced 50% more shoot weight than the highest rate of fertilizer N without compost.

At the low rate of compost application and without fertilizer N addition, uninoculated compost did not increase shoot weight but inoculated compost increased shoot weight 44% (Table 3). Both compost types approximately doubled shoot weight when added at the high rate without fertilizer N in comparison to the treatment not receiving fertilizer N or compost. At the low rate of compost addition and for comparable treatments of fertilizer N, shoot weight was significantly greater for treatments receiving inoculated compost than those receiving uninoculated compost. At the high rate of compost addition and for the two lower levels of fertilizer N addition, shoot weight was also significantly greater for inoculated compost than uninoculated compost. At the highest level of fertilizer N addition and the higher level of compost addition, shoot weight was not significantly different between inoculated and uninoculated compost treatments.

In treatments with compost, root dry weight was significantly influenced by the type of compost and the rate of compost addition but was not significantly influenced by N rate and there were no significant interactions between these factors (Table 2). The root weights for the main effect of compost type were 19.2 g per plant for uninoculated compost and 22.0 g for inoculated compost. The root weights for the main effect of compost rate were 17.6 g per plant for the low level of compost addition and 23.6 for the higher level. When compost was not added, the highest rate of N addition significantly increased root weight (Table 4). The only individual treatment that was significantly different between compost types occurred at the high rate of compost addition. Without fertilizer N, inoculated compost provided significantly higher root weights than uninoculated compost (Table 4).

The main effects of compost type, compost rate and fertilizer N rate influenced the %N in tomato but there were no significant interactions between these factors (Table 2). At the low and high rates of compost addition both composts significantly increased N percentage over no compost addition (Table 5). The N percentage in tomato was higher for inoculated compost than

Table 4. Root weight of tomato as influenced by N rate, compost type, and compost rate

Nitrogen rate	Compost	Compost rate/pot[a]	
		15 g g/plant	45 g
0	None[b]	9.4 c	9.4 d
0.75	None	9.6 be	9.6 d
1.54	None	15.6 ab	15.6 c
0	Uninoculated	17.2 a	19.9 bc
0.75	Uninoculated	16.2 a	23.1 ab
1.54	Uninoculated	17.7 a	21.7 ab
0	Inoculated	16.0 a	26.3 a
0.75	Inoculated	17.8 a	23.8 ab
1.54	Inoculated	21.0 a	27.0 a

[a]Numbers within columns not having letters in common were significantly different at P (0.05).
[b]The treatment not receiving compost was included in the analysis of variance for comparative purposes.

Table 5. Main effect of compost type added at different rates on percentage N in tomato

Compost	Compost rate 1 pot[a]	
	15 g	45 g
None[b]	0.86 a	0.86 a
Uninoculated	1.13 b	1.67 b
Inoculated	1.50 c	1.96 c

[a]Numbers within columns not having letters in common were significantly different at P (0.05).
[b]The treatment not receiving compost was included in the analysis of variance for comparative purposes.

Table 6. Main effect of nitrogen rate on percentage N in tomato receiving different rates of compost

Nitrogen rate/pot	Compost rate/pot		
	0	15 g	45 g
0	0.57[a] a	1.04 a	1.45 a
0.54 g	0.70 a	1.29 b	1.78 b
1.74 g	1.30 b	1.61c	2.21 c

[a]Numbers within columns not having letters in common were significantly different at P (0.05).

Table 7. Populations of rhizobacteria on the roots of tomato as influenced by the quantity of compost added

Genera	Compost rate/pot[a]	
	15 g $\log_{10}$ g^{-1} root	45 g
Beijerinkia	2.51 a	2.62 a
Azotobacter	3.50 b	3.49 b
Azospirillum	4.68 c	4.82 c

[a]Numbers not having letters in common were significantly different at P (0.05).

uninoculated compost treatments. The main effect of fertilizer rate for each rate of compost addition is shown in Table 6. Nitrogen fertilizer addition significantly increased the N percentage in tomato for all rates of compost addition. The highest percentage N was in the treatment receiving the most compost and fertilizer N.

The rhizobacteria colonized roots of plants receiving the inoculated compost (Table 7). At 55 days of growth the populations on tomato roots were highest for *Azospirillum, Azotobacter* and *Beijerinckia*, respectively. Populations on the roots of plants from the treatments receiving uninoculated compost or no compost were below detectable numbers. The higher rate of inoculated compost addition did not significantly increase numbers above that of the lower rate of inoculated compost addition.

Discussion

When no fertilizer N was added, the higher rate (45 g pot^{-1}) of the uninoculated compost doubled shoot growth (Table 3). Inoculated compost increased growth even further (Table 3). The impact of compost on plant growth was most likely due to increasing N availability since it was limiting in this soil. This is supported by the observation that addition of fertilizer N alone increased shoot growth by 40% (Table 3) and adding fertilizer N or compost increased percentage N in plant tissue (Table 6).

Addition of compost provided extra P, K, and Ca besides N (Table 1). However since a relatively large amount of P and K were applied as fertilizer to the soil it is not likely that under these conditions P and K from compost increased plant growth. Although the composts were good sources of Ca the amount contained in the relatively small amount of compost added in comparison to the quantity in 10 kg of soil does not make it likely that it was the reason for the plant growth response to compost addition.

Inoculated compost was clearly superior to uninoculated compost in promoting plant growth (Table 3) and N content (Table 5). Because the N_2 fixing bacteria were on the tomato roots receiving inoculated compost (Table 7) and not at detectable numbers on roots receiving uninoculated compost they appear to mediate a role in increasing tomato growth. Although this study does not allow us to clarify the exact underlying mechanisms for the observed promotion of plant growth a few possibilities can be discussed. Plants inoculated with N_2-fixing bacteria may have increased growth because the bacteria fix N, (Kumar and Narula, 1999; Kumar and Singh, 2001), and/or provide plant growth hormones (Amooaghaie et al., 2002; Bashan and Holguin, 1997; Bashan and Levanomy, 1990; Lin et al., 1983). Additionally our previous research (Meunchang et al., 2005b) had suggested that

inoculated compost resulted in 10% higher rates of organic matter mineralization. If the bacteria were able to increase organic matter mineralization in the soil this also could result in more N being mineralized from the soil thus becoming plant available and perhaps especially important for fast growing plants like tomatoes.

When the growth response from N_2-fixing bacteria is due to plant growth hormones root mass is greatly increased (Vessey, 2003). In our investigation there was a statistically significant increase in root mass due to compost type (Table 2) but the individual treatment comparisons between compost types were not statistically significant (Table 4). From this lack of major response in root growth it is unlikely that the impact of inoculated compost was due to plant growth promoting hormones.

The inoculated compost showed significant acetylene reduction activity during the composting process and up to 16% increase in total N content (Meunchang et al., 2005b) indicating that the inoculant bacteria were capable of N fixation and may have provided some additional N to the plants. However, the role of increased N mineralization in the soil cannot be ruled out in our study and deserves additional study.

In conclusion, the results from investigation suggest that sugar mill wastes composted as a mixture of bagasse and filter cake were effective in increasing tomato growth. Inoculation of the compost with N_2 fixing bacteria provided additional benefit to tomato plants. The use of the waste product from sugar mills as a soil amendment provides affordable fertilizer to farmers and protects the environment, hence turning waste to wealth. Future work needs to be conducted using soil not receiving fertilizer P and plants growing to maturity to more fully evaluate the value of sugar mill waste compost.

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