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RESEARCH ARTICLE

Use of composts to manage corky root disease in organic tomato production

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Abstract

Corky root disease of tomato caused by *Pyrenochaeta lycopersici* is an economically important disease in organic tomato production. This study aimed to evaluate the effects of various composts consisting of green manure, garden waste and horse manure against corky root disease through bioassay under greenhouse conditions, where soil naturally infested with *P. lycopersici* was used as a root substrate. The various composts were mixed at a rate of 20% (v/v) with the infested soil. Disease severity (measured as infected roots) in the unamended soil was compared with that in the soil–compost mixtures. One of the composts made from garden waste significantly reduced the disease, whereas horse manure compost significantly stimulated it. Lower concentrations of NH_4-N and total carbon and a higher concentration of Ca in the substrate were correlated with lower level of corky root disease. Addition of green manure or garden waste compost to the infested soil increased total microbial activity or population density of copiotrophic bacteria and actinomycetes, respectively. However, increased microbial activity or microbial population in soil–compost mixtures was not associated with a reduction in corky root disease severity in the present study.

Introduction

The soil-borne fungal disease corky root of tomato, caused by *Pyrenochaeta lycopersici* Schneider & Gerlach, is a serious problem for tomato production in greenhouses using soil, and also in the field. The disease alters the root system, causing brown lesions on small and larger roots and rotting of small feeder roots (Pohronezny & Volin, 1991). As a result, water and nutrient uptake of infected plants is disturbed, leading to yield losses (Goodenough & Maw, 1973). In greenhouses where the same soil is cultivated continuously for 3–4 years with tomato plants, the disease may cause 30–40% yield reductions and losses of up to 75% have been observed in European greenhouses (Forsberg *et al.*, 1999). The current control methods for corky root disease have various limitations. For example, steaming or solarisation of the soil can reduce the inoculum of corky root fungus in the soil (Last & Ebben, 1968; Moura & Palminha, 1994; Ioannou, 2000). However, soil solarisation is suitable only for countries exhibiting a warm and sunny climate. Steaming is quite costly and there is a risk that the inoculum of *P. lycopersici* will be left in deeper soil layers because of the limit of steam penetration. Steaming may also negatively affect the whole microflora and fauna in the soil. Grafting a commercial cultivar to a rootstock tolerant to *P. lycopersici* is another option but this method is also quite costly. The known source of resistance to corky root disease, the *pyl* gene, exhibits incomplete penetrance and expressivity and thus more research for new genetic resources is still needed (Fiume & Fiume, 2003).

Composts have been used successfully to suppress soil-borne pathogens such as *Fusarium*, *Phytophthora*, *Pythium*, *Rhizoctonia* and *Verticillium* (Chen *et al.*, 1987; Diab *et al.*, 2003; Noble & Coventry, 2005; Termorshuizen *et al.*, 2006). Particle size, electrical conductivity (EC) (soluble salt content), pH, nitrogen content, cellulose and lignin content, and inhibitors released by composts are known physical and chemical properties of composts that affect disease suppression (Hoitink & Fahy, 1986). In composts, total microbial activity, microbial biomass, total number of culturable actinomycetes and the presence of other microorganisms have been suggested to enhance the suppression of various plant diseases (Diab *et al.*, 2003; Noble & Coventry, 2005; Pérez-Piqueres *et al.*, 2006). Field investigations of organic and conventional tomato production systems in the Central Valley of California showed that corky root disease was less prevalent in organic farms than conventional farms (Workneh *et al.*, 1993). It was suggested that the organic farm soil, which was fertilised by various composts, had a higher microbial activity than the conventionally managed soil and that microbial activity was a significant factor associated with lower corky root disease. However, use of compost can also be an interesting approach to corky root disease management in conventional tomato production especially with reduction in available chemicals like methyl bromide. Composts vary considerably in chemical, physical and biotic composition and consequently in ability to suppress soil-borne diseases. From 120 bioassays including 18 composts and 7 pathosystems, significant disease suppression was found in 54% of the cases, while 43% of the cases had no effect and 3% of the cases showed significant disease aggravation (Termorshuizen *et al.*, 2006).

In the present study, the effect of four types of compost on corky root disease of tomato was evaluated in pot experiments under greenhouse conditions. The disease severity in soil naturally infested with *P. lycopersici* was compared with that of compost-amended soil (soil–compost, 80–20% by volume). The relationship between corky root disease severity and several abiotic and biotic properties of the composts was determined. The objectives of this study were (a) to verify the possibility of using composts in corky root disease management and (b) to relate biotic and abiotic properties of the composts to corky root disease severity.

Materials and methods

Soil and composts

Pyrenochaeta lycopersici-infested soil was collected from an organic tomato grower's greenhouse in the vicinity of Uppsala (59°49′N, 17°43′E), Sweden. The infested soil was used for greenhouse experiments in two consecutive years and stored at 4°C between years. In the study, the soils from year 1 and year 2 were referred as infested soil 1 and infested soil 2, respectively. Four types of compost were used (a) green manure compost (GMC), (b) horse manure compost (HMC), (c) garden waste compost 1 (GC1) and (d) garden waste compost 2 (GC2). All composts originated from Sweden. For organic tomato growers in Sweden, it is possible to produce GMC or HMC on farms from locally available resources or buy garden wastes composts from a commercial composting plant. GMC was prepared from red clover harvested at an early stage of development and mixed with chopped straw (10–20% of total dry matter). The compost pile was put outside in summer 2002, with turning once a week during the first month and then every second week. The compost pile was left outside over the winter and later stored frozen (−20°C). The compost sample was taken from the frozen sample early in spring 2004 and stored at 4°C prior to use in summer 2004. GC1 and GC2 were collected from a commercial composting plant in Sala (59°55′N, 16°38′E), Sweden. GC1 consisted of 70% (vol.) garden waste and 30% horse manure with an unknown proportion of straw. GC2 consisted of 70% (vol.) garden waste and 30% horse manure with an unknown proportion of peat. The wastes were crushed with a high-speed shredder and put with straw or peat in an uncovered windrow. The materials were turned once a week with a windrow turner for 3 months. To encourage further decomposition, all materials were placed outside in a large heap measuring 3 × 15 × 15 m (h × w × l) and the heap was turned once a month for 6 months. The heap age was 1.6 years for GC1 and 1.0 year for GC2 when compost samples were collected for our experiments. HMC, consisting of unknown proportions of horse manure and peat, was collected in spring 2005 from a horse manure pit in Uppsala where the mixture had been composted for 8–9 months. During collection of compost samples, random samples were taken from different places in the compost heap and these subsamples were mixed together. The composts were stored at 4°C as the infested soil until used.

Chemical properties of soil, composts and soil–compost mixtures

EC, pH and readily available NO_3-N, NH_4-N, B, Ca, Cl, K, Mg, Mn, Na, P, S of the soil, composts and soil–compost mixtures were determined by methods described by Spurway & Lawton (1949) in a soil analysis laboratory, Lennart Månsson International (LMI), Helsingborg, Sweden. Analysis of total contents of different plant nutrients in the soil, composts and soil–compost mixtures was performed by the Department of Soil Science, Swedish

University of Agricultural Sciences, Uppsala, Sweden. Total carbon (tot-C) and N were determined after dry combustion on an auto-analyser (LECO CNS 2000, LECO Corporation, St Joseph, MI, USA). Total NH_4-N and NO_3-N were determined colorimetrically by an auto-analyser TRAACS 800 (Bran & Lubbe, Hamburg, Germany) after extraction with 2 M KCl. Total contents of B, Ca, Fe, K, Mg, Mn, Na, P and S were determined with an inductively coupled plasma emission spectrometer (ICP, Perkin Elmer Optima 3000, Petkin Elmer, Norwalk, CT, USA) after dissolution in concentrated HNO_3. The chemical properties of soil, compost and soil–compost mixtures are presented in Table 1 (according to the analysis of total nutrient contents of the substrates).

Biological properties of soil, composts and soil–compost mixtures

Total culturable copiotrophic and oligotrophic bacteria, fungi and actinomycetes were determined by the dilution-plating technique as described by Termorshuizen *et al.* (2006). Samples of 10 g (d.wt basis) substrate were suspended in 90 mL water, sonicated for 1 min and shaken (180 r.p.m., 30 min). A serial 10-fold dilution from 10^{-1} to 10^{-8} was prepared. Plating was performed from dilutions 10^{-4}, 10^{-5}, 10^{-6}, 10^{-7} and 10^{-8} for copiotrophic bacteria, 10^{-2}, 10^{-3} and 10^{-4} for oligotrophic bacteria and actinomycetes, and 10^{-3}, 10^{-4} and 10^{-5} for fungi. 100-μl aliquots of each dilution were pipetted onto agar plates. For copiotrophic bacteria, high nutrient medium was used: 0.5 g $MgSO_4 \cdot 7H_2O$, 0.5 g KNO_3, 1.3 g $K_2HPO_4 \cdot 3H_2O$, 0.06 g $Ca(NO_3)_2 \cdot 4H_2O$, 2.5 g $C_6H_{12}O_6$, 0.2 g enzymatic casein hydrolysate, 15 g Technical agar (Oxoid no. 3; Oxoid, Hampshire, UK), 1 L demineralised water and 100 mg L^{-1} cycloheximide after sterilisation. For oligotrophic bacteria and actinomycetes, the same medium was used at 100-fold dilution of all ingredients except the agar where 15 g L^{-1} Agar Noble (Difco, Detroit, MI, USA) was used instead of Technical agar. After sterilisation, 100 mg L^{-1} cycloheximide was also added. For fungal medium, 50 g L^{-1} malt extract (Oxoid) and 15 g L^{-1} agar–agar (Merck, Darmstadt, Germany) was amended with 50 mg L^{-1} oxytetracycline dihydrate (Sigma, St Louis, MO, USA) after sterilisation. Three plates per dilution were used. Inoculated plates were incubated in darkness at 25°C for 7 days (copiotrophic bacteria and fungi) or 28 days (oligotrophic bacteria and actinomycetes). Colonies were enumerated and populations were expressed as the number of colony forming units per gram dry weight of the substrate.

Total microbial activity of soil, compost and soil–compost mixtures was measured by determining basal respiration (CO_2 evolution rate) in a glass jar containing one plastic vial (length 7 cm, diameter 2.5 cm) containing compost (2 g) or soil–compost mixture (10 g) and two plastic vials (length 6 cm, diameter 1 cm) containing 4 mL NaOH (0.2 M) each. The glass jar was incubated at 20°C for 48 h. In this system, the CO_2 produced was trapped in NaOH solution and was measured by means of titration against HCl (0.1 M). Basal respiration was expressed as mg CO_2 g^{-1} d.wt day^{-1}. The determination was performed in triplicate for soil, composts and soil–compost mixtures.

Table 1 Chemical properties of the soil, compost and soil–compost mixtures used

	IS1[a]	GMC	GC2	IS1 + GMC	IS1 + GC2	IS2[a]	HMC	GC1	IS2 + HMC	IS2 + GC1
pH	6.7	10.5	7.7	7.7	6.9	7.1	6.7	8.1	6.7	7.0
EC	7.6	27.9	2.4	9.7	4.6	7.7	5.6	7.5	6.7	7.1
Tot-C (% of d.wt)	12.7	35.7	28.4	13.6	10.5	11.7	39.9	17.5	12.8	9.4
Tot-N (% of d.wt)	1.01	4.1	1.5	0.8	0.7	0.9	1.8	1.8	0.8	0.9
C : N	–	8.7	19.2	–	–	–	22.5	9.8	–	–
NH_4-N (mg kg^{-1} d.wt)	6.5	191	2.4	44.7	1.6	3.3	143.6	3.4	178.1	2.7
NO_3-N (mg kg^{-1} d.wt)	130.9	2890.3	100.5	463.1	179.0	212.2	127.7	57.5	224.5	132.5
$NH_4 : NO_3$	–	0.07	0.02	–	–	–	1.12	0.06	–	–
P (g kg^{-1} d.wt)	4.0	5.5	2.7	2.2	2.6	2.2	2.9	2.6	2.5	2.5
K (g kg^{-1} d.wt)	6.9	74.3	9.4	10.7	5.6	7.3	24.4	8.3	9.1	6.8
Na (g kg^{-1} d.wt)	1.2	0.2	0.6	0.9	0.8	0.9	2.0	2.3	0.9	1.0
B (g kg^{-1} d.wt)	0.02	0.06	0.02	0.01	0.01	0.01	0.04	0.02	0.01	0.04
Ca (g kg^{-1} d.wt)	11.8	30.9	16.5	18.9	14.7	15.8	8.7	60.6	12.5	19.6
Fe (g kg^{-1} d.wt)	20.9	2.0	12.0	17.1	20.4	25.6	1.5	8.7	23.1	19.1
Mg (g kg^{-1} d.wt)	7.4	8.8	5.6	6.5	8.2	8.1	2.7	15.2	7.4	7.5
Mn (g kg^{-1} d.wt)	0.5	0.1	0.4	0.6	0.5	0.7	0.1	0.3	0.7	0.6
S (g kg^{-1} d.wt)	2.1	4.2	3.4	2.3	1.8	1.6	3.2	3.2	1.8	3.3

EC, electrical conductivity; GC1, garden waste compost 1; GC2, garden waste compost 2; GMC, green manure compost; HMC, horse manure compost; IS1, infested soil 1; IS2, infested soil 2; Tot-C, total carbon; Tot-N, total nitrogen. –, Not calculated.

[a] The infested soil was referred as IS1 and IS2 as the soil was used in two consecutive years.

To determine the initial nematodes population in the soil, composts and soil–compost mixtures, nematodes were extracted by the Baermann funnel method (Southey, 1986) for 24 h at approximately 20°C, with four 30 mL replicates for each substrate. Nematodes were counted under a stereomicroscope at 25–50× magnification and categorised into one of the two groups: (a) fungivorous nematodes or (b) bacteriovorous and other (non-fungivorous) nematodes.

Sterilisation of the infested soil

To determine whether the brown discoloration of roots observed was caused by corky root disease, the infested soil was sterilised by 2.5 Mrad of gamma radiation (CODAN Steritex ApS, Espergaerde, Denmark) to eliminate *P. lycopersici*. The irradiated soil was incubated at room temperature (20–22°C) for 5 weeks prior to use.

Greenhouse experiments

Three greenhouse experiments were conducted following a fully randomised design. For the treatments, there were 10 replicates in experiment 1 and eight replicates in experiments 2 and 3. An overview of greenhouse experiments is presented in Table 2.

The infested soil and composts were incubated for 1 week at room temperature (20–22°C) prior to greenhouse experiments. Composts (20% vol.) were mixed with the soil by a rotary shaker. Perlite (Norwegian Perlite®; Norwegian Perlite, Sande, Norway; size: 1.5–6.0 mm) was added to the unamended soil (20% vol.) to compensate for the compost by volume. Equivalent inorganic nutrients of 20% compost (according to the analysis by the Spurway & Lawton method) were prepared by dissolving different salts in distilled water and added to the infested soil–perlite substrate. Equivalent inorganic nutrients of 20% GMC, HMC, GC1 and GC2 were added to the irradiated and non-radiated infested soil and a bioassay was performed to determine the disease severity in the respective soils. The pH of the nutrient solution was adjusted to that of the corresponding soil before application.

Tomato seedlings were prepared by sowing seeds (cv. Elin, Weibulls®; Weibulls, Svalöv, Sweden) in commercial soil (Hasselfors Garden E-Jord®, Hasselfors Garden, Örebro, Sweden). Three-week-old seedlings were transferred to plastic pots with 5 L substrate in the greenhouse containing a single seedling in each pot. The average temperature during the experiments varied from 18 to 22°C and relative humidity was kept at 70%. Artificial light was used only in experiment 2, for 16 h per day for 1 month before harvesting, because of short day length. The plants were irrigated twice a day with normal tap water. To avoid nutrient deficiency, organic fertiliser was applied to all treatments when the plants were 8 weeks old.

Greenhouse experiments continued for 10 weeks after transplanting of tomato seedlings, because an earlier trial in this greenhouse on transplanting 3-week old tomato seedlings in pots with 5 L infested soil showed that the plants required 10 weeks to develop characteristic corky root disease symptoms on roots. During the experiment, fruits were picked as they ripened. At harvest, all fruits (green and red fruits) from each plant were weighed together and total fruit weight was expressed as cumulative weight. The shoots were cut-off at the soil surface. The roots were separated from the soil by gentle shaking and rinsed with tap water to remove soil particles. Disease severity in each plant was evaluated by collecting the following three 3-cm sections of root sample: leaving a segment of 5 cm from the root base and then taking a 3-cm sample, leaving 5 cm and then taking another 3-cm

Table 2 Design of the greenhouse experiments studying the effect of composts on corky root disease in tomato plants grown in GMC, garden waste compost and HMC mixed with soil naturally infested by *Pyrenochaeta lycopersici*

Experiments	Treatments
Experiment 1	IS1[a]
	IS1 + GMC (20% by volume)
	IS1 + perlite (20% by volume)
	IS1 + perlite (20% by volume) + Nut.GMC
Experiment 2	IS1
	IS1 + GC2 (20% by volume)
	IS1 + perlite (20% by volume)
	IS1 + perlite (20% by volume) + Nut.GC2
Experiment 3	IS2[a]
	IS2 + HMC (20% by volume)
	IS2 + GC1 (20% by volume)
	IS2 + perlite (20% by volume)
	Irradiated IS2
	IS2 + perlite + Nut.GMC
	Irradiated IS2 + perlite + Nut.GMC
	IS2 + perlite + Nut.GC2
	Irradiated IS2 + perlite + Nut.GC2
	IS2 + perlite + Nut.HMC
	Irradiated IS2 + perlite + Nut.HMC
	IS2 + perlite + Nut.GC1
	Irradiated IS2 + perlite + Nut.GC1

GC1, garden waste compost 1; GC2, garden waste compost 2; GMC, green manure compost; HMC, horse manure compost; IS1, infested soil 1; IS2, infested soil 2; Nut.GC1, equivalent inorganic nutrients of 20% garden waste compost 1; Nut.GC2, equivalent inorganic nutrients of 20% garden waste compost 2; Nut.GMC, equivalent inorganic nutrients of 20% green manure compost; Nut.HMC, equivalent inorganic nutrients of 20% horse manure compost.

[a] The infested soil was referred as IS1 and IS2 as the soil was used in two consecutive years.

sample, leaving 5 cm and then taking another 3-cm sample. Then the three root samples from three distances of each plant were pooled and mixed. From these root samples, 100 pieces from each plant were examined under a stereomicroscope and grouped into three categories as white (healthy root), light brown (initially infected root) and dark brown (severely infected root).

Statistical analysis

To model the probabilities of healthy, initially infected and severely infected roots, a generalised linear model for ordinal-scaled observations was fitted with the procedure GENMOD in SAS. The logit link was used and overdispersion within the root was modelled with the option DSCALE. For disease severity in different treatments, the analysis was made with the treatments as explanatory factors. CONTRASTS were used to separate different treatments. For the relationship between corky root disease severity and biotic and abiotic properties of soil and soil–compost mixtures, the analysis was made with the properties as continuous explanatory variables. Microbial population densities (colony numbers of copiotrophic and oligotrophic bacteria, actinomycetes and fungi), nematode numbers and basal respiration were analysed by ANOVA in Minitab (version 14) and treatment differences were compared by least significant difference test at $P \leq 0.05$. Data on microbial population densities and nematode numbers were log transformed prior to analysis. The physical and chemical properties of the soil and composts were submitted to principal component analysis (PCA) in Minitab (version 14).

Results

Effect of composts on corky root disease

Corky root disease severity was reduced significantly ($P < 0.0001$) by 13% in the GC1 treatment. In contrast, significant 19% disease aggravation ($P < 0.0001$) was observed in the HMC (Fig. 1). The two other composts, GMC and GC2, had no significant effect on corky root disease (Figs 2 and 3).

Chemical properties of soil and composts

The soil and the composts tested had different physiochemical properties, leading to a clear distinction between them after PCA analysis (Fig. 4). There was an indication that GMC was high in pH, EC values and NO_3-N content, while GC1 was high in Mg and Ca content and low in tot-C content. HMC was low in Mg and Ca content. GC2 and the infested soils were high in Fe and Mn content.

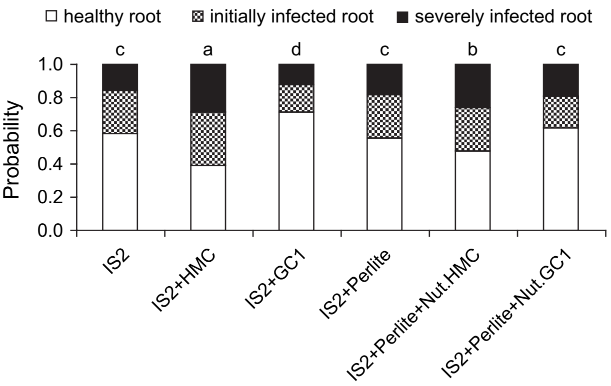

Figure 1 Effect on corky root disease of horse manure compost (HMC) and garden waste compost 1 (GC1) and equivalent inorganic nutrients of 20% horse manure compost (Nut.HMC) and garden waste compost 1 (Nut.GC1) in the infested soil 2 (IS2). Different small letters on top of the bars indicate significant differences ($P \leq 0.05$).

Biological properties of soil, composts and soil–compost mixtures

Addition of GMC, GC2, GC1 and HMC in the infested soil significantly ($P \leq 0.05$) increased the number of copiotrophic bacteria but not oligotrophic bacteria. Only GC1 significantly ($P \leq 0.05$) increased the number of actinomycetes in the infested soil. Fungal population density in the infested soil was not altered by the addition of any of the composts. The numbers of fungivorous and bacteriovorous and other (non-fungivorous) nematodes decreased

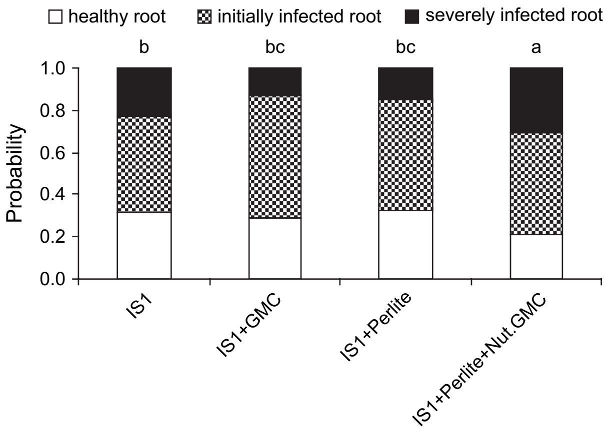

Figure 2 Effect on corky root disease of green manure compost (GMC) and equivalent inorganic nutrients of 20% green manure compost (Nut.GMC) in the infested soil 1 (IS1). Different small letters on top of the bars indicate significant differences ($P \leq 0.05$).

significantly ($P \leq 0.05$) after addition of GMC. Microbial activity (measured as basal respiration) of the infested soil was increased significantly ($P \leq 0.05$) by the addition of GMC but not by the other three composts (Table 3).

Relationship between chemical and biological properties of soil and soil–compost mixtures and disease severity

There were significant correlations between ammonium nitrogen (NH_4-N), tot-C and calcium (Ca) concentration in the substrate and corky root disease severity (Fig. 5). Disease severity increased significantly with increasing concentration of ammonium nitrogen ($P = 0.05$) and tot-C ($P < 0.0001$) in the substrate, while the disease severity was significantly reduced by an increased concentration of calcium ($P < 0.0001$). No significant relationship was found between biological properties of the substrate and severity of corky root diseases.

Effect of inorganic nutrients on corky root disease

Addition of equivalent inorganic nutrients of 20% garden waste compost 1 (Nut.GC1) and green manure compost (Nut.GMC) significantly increased disease severity ($P < 0.0001$) in the infested soil compared with the respective GC1-amended and GMC-amended soil (Figs 1 and 2). Addition of equivalent inorganic nutrients of 20% horse manure compost (Nut.HMC) significantly decreased disease severity ($P = 0.0057$) in the infested soil compared with HMC-amended soil (Fig. 1), whereas inorganic nutrients of garden waste compost 2 (Nut.GC2) had no effect on disease (Fig. 3).

In irradiated infested soil, inorganic nutrients of four composts caused almost no brown roots. Conversely, inorganic nutrients of four composts caused more brown roots in non-irradiated infested soil (Fig. 6).

Effect of composts on plant growth

Addition of any of the four composts did not significantly increase shoot and root dry weight and total fruit weight (data not shown).

Discussion

Compost amendment had variable effect on corky root disease severity: addition of GC1 to the infested soil reduced corky root disease, whereas addition of HMC increased the disease and the two other composts had no effect. This variable effect is in line with the study by Termorshuizen *et al.* (2006), who observed different responses of plant pathogens to different composts. The effect of different composts on a pathogen may vary as compost characteristics, both physiochemical and biological, vary between different composts.

Ammonium nitrogen has been shown to exacerbate several root rots and cortical rot diseases of plants caused by *Fusarium*, *Phytophthora* and *Rhizoctonia* (Huber & Watson, 1974; Nasir *et al.*, 2003). In the current study, increase in concentration of NH_4-N was associated with increase in corky root disease severity. Thus, the lower amounts of NH_4-N in GC1-amended soil might be a reason for the lower severity of disease in this soil. Similarly, higher amounts of NH_4-N in HMC-amended soil than all other three compost-amended soils might have caused higher disease in this soil. However, the concentration of NO_3-N

Table 3 Biological properties of compost-amended and non-amended soils used in the study[a]

	CFU ($\times 10^5$ g^{-1} Dry Substrate)				Nematode Numbers mL^{-1} Substrate		
Growth Medium	Copiotrophic Bacteria	Oligotrophic Bacteria	Actinomycetes	Fungi	Fungivorous	Bacteriovorous and Other (Non-Fungivorous) Nematodes	Basal Respiration (mg CO$_2$ g^{-1} Dry Substrate day^{-1})
IS1	4.86 b	4.70 a	1.12 a	0.16 a	0.80 a	11.75 a	0.18 b
IS1 + GMC	7.36 a	5.03 a	1.46 a	0.19 a	0.16 b	6.27 b	0.58 a
IS1 + GC2	7.20 a	5.13 a	1.67 a	0.16 a	0.61 a	13.03 a	0.37 ab
LSD 5%	2.25	2.92	1.50	0.07	0.33	4.87	0.34
IS2	4.12 b	4.32 a	1.13 b	0.16 a	0.69 a	7.85 a	0.16 a
IS2 + HMC	7.13 a	4.61 a	1.30 ab	0.17 a	0.43 a	9.01 a	0.33 a
IS2 + GC1	7.00 a	4.93 a	2.23 a	0.15 a	0.55 a	14.32 a	0.24 a
LSD 5%	1.93	2.04	0.94	0.11	0.64	13.83	0.18

CFU, colony forming units; GC1, garden waste compost 1; GC2, garden waste compost 2; GMC, green manure compost; HMC, horse manure compost; IS1, infested soil 1; IS2, infested soil 2; LSD, least significant difference.

[a] Data are mean values, where $n = 3$ for population densities of copiotrophic bacteria, oligotrophic bacteria, actinomycetes, fungi and for basal respiration and $n = 4$ for nematode numbers. Within each column of IS1, IS1 + GMC, IS1 + GC2 and IS2, IS2 + HMC, IS2 + GC1, values followed by different letters are significantly different (LSD test, $P \leq 0.05$).

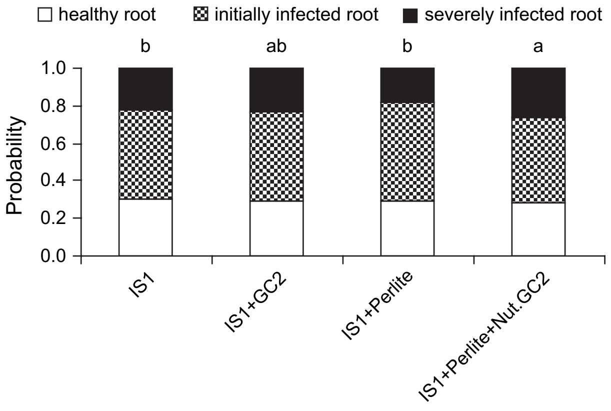

Figure 3 Effect on corky root disease of garden waste compost 2 (GC2) and equivalent inorganic nutrients of 20% garden waste compost 2 (Nut.GC2) in the infested soil 1 (IS1). Different small letters on top of the bars indicate significant differences ($P \leq 0.05$).

in the substrate was not related to disease severity in the present study. In general, the form of nitrogen may influence the virulence of a pathogen by affecting extracellular enzyme production, and thus inhibition or activation of enzyme synthesis could account for the disease severity (Huber & Watson, 1974). In addition to NH_4-N, GC1-amended soil had the lowest content of tot-C, which meant the energy available for microorganisms was relatively limited that may have led to a relatively high level of microbial competition. *P. lycopersici* is most likely suppressed in such a highly competitive situation, as it has a low competitive ability (Davet, 1976). Competition between *P. lycopersici* and other microorganisms in soils has been suggested as a mechanism of corky root disease suppression (Workneh & Bruggen, 1994*a*).

The highest concentration of calcium found in GC1 and in soil amended with this compost might be another reason for the disease reduction observed. Calcium is an important element in protecting plants against certain cell-wall degrading enzymes produced by fungal pathogens (Conway & Sams, 1984). Calcium treatment (as $CaCO_3$) of tomato plants reduced *Fusarium* crown rot disease (Woltz *et al.*, 1992). In our study, we also found that corky root disease severity decreased with increased concentration of calcium in the substrate. Therefore, the lower concentration of calcium found in HMC-amended soil might be another reason for the higher disease severity there. However, the application of Ca to soil has never been tested directly with respect to *P. lycopersici*. This can be interesting in future study.

Addition of Nut.GMC and Nut.GC1 caused higher disease severity in the infested soil compared with the respective compost-amended soils. In the soil where equivalent inorganic nutrients were added, biotic properties of the composts were absent. Higher disease severity in the absence of biotic properties of the composts is an indication of the involvement of biological properties of these two composts in disease suppression. However, GMC did not reduce disease severity in the present study, although addition of this compost to the infested soil did increase total microbial activity significantly. In GMC-amended soil, the ammonium level was higher than in the two garden waste compost-amended soils. Thus, disease reduction by high microbial activity in GMC-amended soil was probably counteracted by the high ammonium level.

Generally, addition of compost to the main substrate increases the abundance of microbial populations in the substrate (Mabuhay *et al.*, 2006; Pérez-Piqueres *et al.*, 2006). In our study, addition of all composts increased the number of copiotrophic bacteria (fast-growing species, r strategists) in the infested soil but not the oligotrophic bacteria (slow growing species, k strategists). A probable reason could be the characteristic of copiotrophic group in responding quickly to excess nutrients, a situation occurring after the addition of composts. The

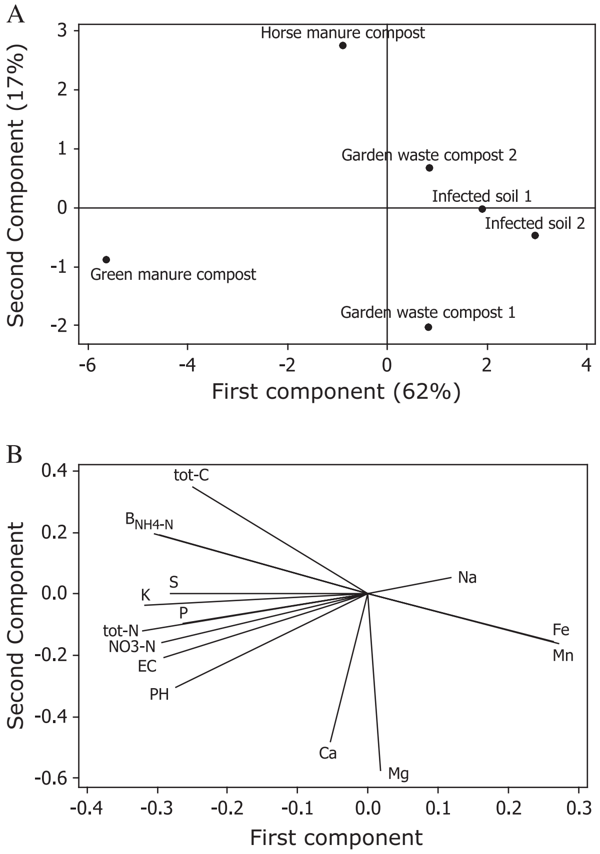

Figure 4 (A) Principal component analysis (PCA) of physical and chemical properties of soil and different composts used in the study. (B) Variables explaining the variation of soil and different composts.

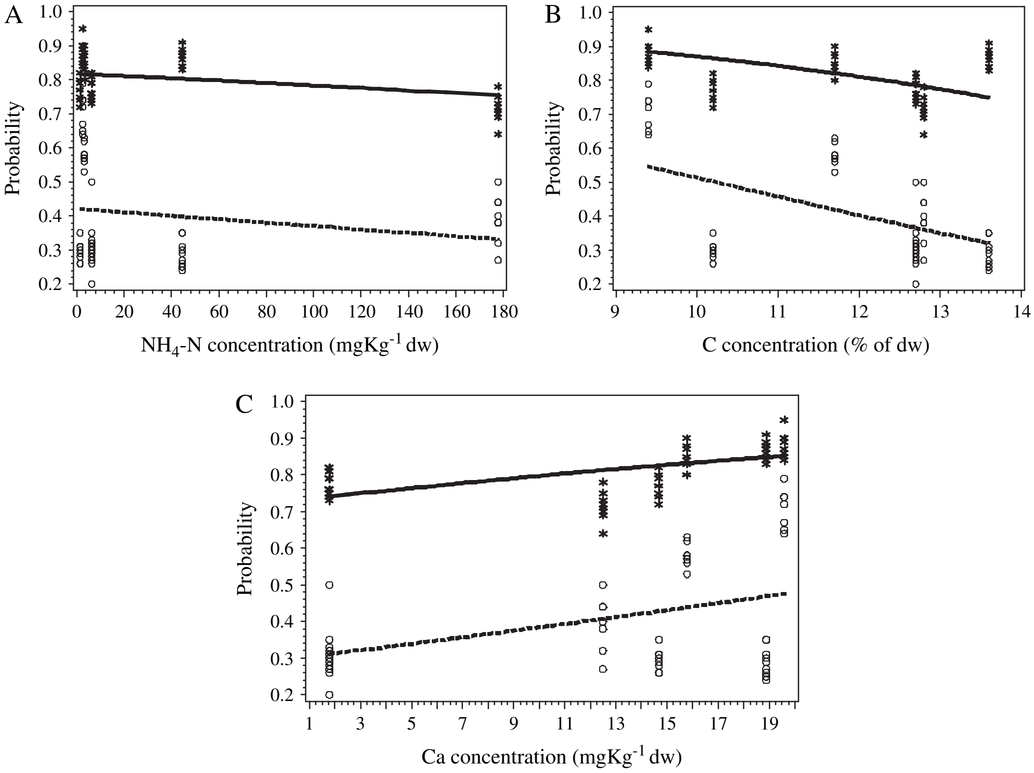

Figure 5 Corky root disease severity as related to (A) NH$_4$-N concentration, (B) C concentration and (C) Ca concentration of soil and soil–compost mixture. Dashed line represents the probability of healthy root and solid line represents the probability of healthy root and initially infected root.

number of actinomycetes in GC1-amended soil was significantly higher than in non-amended soil. This was because of the fact that the compost originated from plant materials, as it has been reported that actinomycetes are abundant in plant residue composts (Crawford, 1988). The number of actinomycetes usually increases in the latter stage of composting, when the remaining substrates of the compost are predominantly cellulose and lignin (Tiquia *et al.*, 2002). GC1 was older than GC2 and, probably for this reason, GC1 had a higher number of actinomycetes than GC2. In a study, corky root disease reduction was positively correlated with the total number of fluorescent *Pseudomonas* (a copiotrophic group) and cellulolytic actinomycetes in the rhizosphere of tomato plants (Workneh & Bruggen, 1994*b*). Actinomycetes have also been associated with suppression of several other plant diseases (Lechevalier, 1988). However, in the present study, we cannot say that the increased number of copiotrophic bacteria and actinomycetes caused by the addition of GC1 was responsible for the disease reduction by this compost because no significant relationship was found between disease reduction and copiotrophic bacteria or actinomycetes. It might be that these two biological components had an influence on *P. lycopersici* that could not be determined during the course of the present greenhouse experiment.

Equivalent inorganic nutrients of 20% composts caused disease in the non-irradiated infested soil, whereas there was no disease when the same nutrients were added to the irradiated soil. This suggests that large amounts of brown roots, which were caused by the addition of inorganic nutrients of GMC and GC1, were because of the infection by *P. lycopersici* and that the root discoloration was not a nutrient burn effect. Brown discoloration on tomato roots may also be caused by *Fusarium oxysporum* f.sp. *radicis-lycopersici* (Shishkoff & Campbell, 1990). However, *Fusarium* was not found in our attempt to isolate this fungus from the current infested soil on synthetic meagre agar (SNA = synthetischer nährstoffarmer agar, Nirenberg) medium, a suitable medium for *Fusarium* identification (Gams *et al.*, 1987). Therefore, we consider that the brown discoloration was caused by *P. lycopersici*. This assumption was strengthened by successful isolation of *P. lycopersici* from brown roots of infested plants on tomato juice agar medium, modifying V8 juice agar medium (McGrath & Campbell, 1983).

The number of fungivorous and bacteriovorous nematodes decreased when GMC was added, probably because

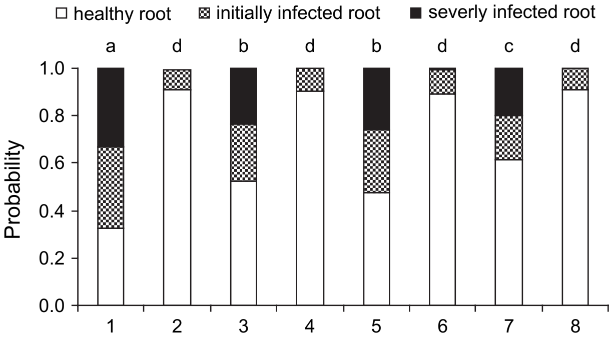

Figure 6 Effect on corky root disease of equivalent inorganic nutrients of 20% composts in non-irradiated infested soil and irradiated infested soil. Different small letters on top of the bars indicate significant differences ($P \leq 0.05$). 1, infested soil 2 (IS2) + perlite + nutrients of 20% green manure compost (Nut.GMC); 2, irradiated infested soil + perlite + Nut.GMC; 3, IS2 + perlite + nutrients of 20% garden waste compost 2 (Nut.GC2); 4, irradiated infested soil + perlite + Nut.GC2; 5, IS2 + perlite + nutrients of 20% horse manure compost (Nut.HMC); 6, irradiated infested soil + perlite + Nut.HMC; 7, IS2 + perlite + nutrients of 20% garden waste compost 1 (Nut.GC1); 8, irradiated infested soil + perlite + Nut.GC1.

of the high EC value in this compost. It is well known that a direct osmotic effect on nematodes because of high salinity can decrease the number of nematodes. For instance, in a study by Oka & Yermiyahu (2002), the high EC value of cattle manure compost caused the suppression of *Meloidogyne javanica* in tomato.

Addition of composts to the infested soil did not improve shoot and root weight and total fruit weight in the present study. This could be because of the short experimental period and early harvesting.

In conclusion, our results show that the nutrient concentration of composts is associated with corky root disease. Low concentrations of NH_4-N and C and high concentrations of Ca reduced corky root disease and therefore the application of composts with such properties may be effective for corky root disease management in tomato.

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