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Influence of soil organic amendments on suppression of the burrowing nematode, *Radopholus similis*, on the growth of bananas

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Abstract *Radopholus similis* is a major constraint to banana production in Australia and growers have relied on nematicides to manage production losses. The use of organic amendments is one method that may reduce the need for nematicides, but there is limited knowledge of the influence of organic amendments on endo-migratory nematodes, such as *R. similis*. Nine different amendments, namely, mill mud, mill ash, biosolids, municipal waste compost, banana residue, grass hay, legume hay, molasses and calcium silicate were applied to the three major soil types of the wet tropics region used for banana production. The nutrient content of the amendments was also determined. Banana plants were inoculated with *R. similis* and grown in the soil-amendment mix for 12-weeks in a glasshouse experiment. Assessments of plant growth, plant-parasitic nematodes and soil nematode community characteristics were made at the termination of the experiment. Significant suppression of plant-parasitic nematodes occurred in soils amended with legume hay, grass hay, banana residue and mill mud relative to untreated soil. These amendments were found to have the highest N and C content. The application of banana residue and mill mud significantly increased shoot dry weight at the termination of the experiment relative to untreated soil. Furthermore, the applications of banana residue, grass hay, mill mud and municipal waste compost increased the potential for suppression of plant-parasitic nematodes through antagonistic activity. The application of amendments that are high in C and N appeared to be able to induce suppression of plant-parasitic nematodes in bananas, by developing a more favourable environment for antagonistic organisms.

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Introduction

The Australian banana industry is composed of subtropical and tropical components. However, the wet tropics region of north Queensland is the most important production area with 88% of the country's banana production (Collins et al. 2004). The wet tropics is located in a region receiving an average of 3 800 mm of rainfall annually, often in heavy downpours. The high rainfall and close proximity to world heritage listed rainforest and coral reefs make the area an environmentally sensitive zone for agriculture. Therefore, environmentally responsible farming practices are required to ensure grower profitability and sustainable management of nutrients, pests and diseases, in particular *Radopholus similis*.

R. similis is a migratory endo-parasitic nematode that feeds on the root cortical tissue of bananas forming dark red lesions, which result in reduced bunch weights, increased vegetative cycling periods and may cause the plant to topple (Gowen et al. 2005). *R. similis* is recognised as the most economically important plant-parasitic nematode on banana world wide (Gowen et al. 2005). To prevent losses caused by *R. similis* in banana production in Australia, banana farmers apply organophosphate or carbamate nematicides, which cost AU$10-15 million annually (Pattison 1994). Nematicides tend to be non-selective biocides, which impact on soil biodiversity and biological processes (Eisenhauer et al. 2010). Also, the application of nematicides is potentially hazardous to the local environment as they can be readily transported in soil water and attached to colloidal soil particles (Cáceres et al. 2002). Furthermore, concerns for worker safety have meant that alternative methods for managing plant parasitic nematodes on bananas are required.

Globally there are a number of research efforts to reduce nematicide use and the impact of plant-parasitic nematodes on banana production, by developing nematode suppressive cropping systems (Gowen et al. 2005). The approach taken is dependent on the technology and resources available and the intensity of banana production. In African subsistence style banana production nematode suppressive systems are being developed using clean planting material, biological antagonists and management of planting configuration (Coyne et al. 2010; Gaidashova et al. 2009; Paparu et al. 2009). For export banana production in Central America and the Caribbean, a greater understanding of how nematodes interact with their hosts and the design of more suppressive systems including weed control is being investigated (Araya 2005; Chabrier and Queneherve 2003; Chabrier et al. 2010; Quénéhervé et al. 2006; Tixier et al. 2006).

The application of organic amendments to soil is viewed as an environmentally favourable waste management strategy that can also potentially improve soil quality (Flavel and Murphy 2006). Considerable research has shown the benefits of using composts and other organic amendments to improve soil physical (water holding capacity, porosity and bulk density), chemical (pH, electrical conductivity and nutrient content) and biological properties such as soil microbial populations and plant growth (Flavel and Murphy 2006; Kennedy et al. 2004; Moss et al. 2002). Amendments have also been investigated for the suppression of soil borne diseases in a range of cropping systems (Stone et al. 2004). Furthermore, there have been investigations to determine the usefulness of soil amendments to reduce plant-parasitic nematode populations in agricultural crops (Akhtar and Malik 2000; Stirling et al. 2003; Widmer et al. 2002).

The results from the application of organic amendments for nematode suppression are variable and sometimes crop, soil or site specific. Amendments may need to be applied continuously at high rates to reduce plant-parasitic nematode populations (Vawdrey and Stirling 1997), which may impact on physical, chemical and biological soil properties.

The chemical composition of the amendments influences the rate of microbial decomposition of organic matter, which can regulate the stability and release of nutrients (Moss et al. 2002). Easily degraded forms of C are preferentially utilised by bacterial populations, while amendments with a high C:N ratio are less rapidly degraded and may stimulate fungal activity (Flavel and Murphy 2006; Valenzuela-Solano and Crohn 2006). Furthermore, the rate of nutrient release may impact on the amendment's potential to stimulate organisms that are antagonistic to plant-parasitic nematodes. It has also been suggested that the inherent qualities of soils has a major impact on the potential to suppress plant-parasitic nematodes (Westphal 2005; Widmer et al. 2002). All agricultural soils are expected to have some capacity to suppress plant diseases depending on their management, with the level of antagonistic potential driven by both biotic and abiotic factors in the soil environment (Sikora 1992; Weller et al. 2002)

An integrated nematode management system has been developed for the Australian banana industry to reduce losses caused by *R. similis* (Stirling and Pattison 2008). However, further work is required to improve the resilience of the banana production system to damage by plant-parasitic nematodes, particularly *R. similis*. The objectives of the following investigations were to examine various soil amendments readily available to banana growers in the wet tropics region of Australia to determine which amendments were able to suppress *R. similis* on bananas and what changes in soil properties were associated with nematode suppression in a glasshouse experiment.

Materials and methods

Soils

Three soils, representative of major soil types for banana production in the wet tropics region of north Queensland, were chosen for a pot experiment; Mundoo, Innisfail and Coom soils series (Table 1). The Mundoo soil is a red uniform, clay loam, well structured Ferrosol soil, with good drainage, derived from basalt (Cannon et al. 1992). The Innisfail soil was a well structured brown clay loam and the Coom soil was a poorly structured, mottled uniform clay loam to clay soil with a silty texture and poor drainage, both being Dermosols (Cannon et al. 1992). All soils used in the experiment were collected from the top 15 cm of commercial banana farms, prior to addition of amendments (Table 1).

Amendments and nematode inoculation

Nine different amendments that were readily available to banana growers in north Queensland were used in the experiment; mill mud, mill ash, molasses (all by-products from sugarcane processing), biosolids, municipal waste compost

Table 1 Location, classification, pH, sand, silt and clay content of typical Australian banana producing soils used in the pot experiment to determine the effect of different soil amendments

Soil series	Classification	Latitude (S)	Longitude (E)	pH	Sand%	Silt%	Clay%
Coom	Dermosol	17°40′58″	146° 3′38″	4.7	5	44	50
Innisfail	Dermosol	17°58′ 9″	146°49′50″	5.3	35	25	40
Mundoo	Ferrosol	17°36′14″	146° 0′41″	4.9	49	10	41

(MW compost), banana residue (*Musa* AAA, Cavendish subgroup), grass hay (*Chloris gayana*), legume hay (*Medicago sativa),* and calcium silicate ($CaSiO_3$). The amendments were mixed with the three different soils and compared to untreated soil. The amendments were analysed for their chemical composition, listed in Table 2, using the methods described by Rayment and Higginson (1992). The biosolids, compost, mill mud, banana residue, grass hay and legume hay were mixed at an air dried rate equivalent to 40 t ha^{-1} (228 g pot^{-1} w/v) (Table 2). Two rates of air dried mill ash were incorporated with the soil, 40 and 120 t ha^{-1} (228 and 678 g pot^{-1} w/v respectively). Furthermore, $CaSiO_3$ was applied at 5 t ha^{-1} (28.2 g pot^{-1} w/v) and molasses at 300 L ha^{-1} (995 mL pot^{-1} v/v). The rate of amendment applied per pot was calculated using the surface area of the 200 mm diameter pots with a depth of 180 mm. The amendments were incorporated and thoroughly mixed with the soil prior to placement in the pots. Approximately 3 kg of the soil and amendment mix was placed in each pot. The untreated soil from each soil type was used as a reference for any changes in soil characteristics as it underwent the same mixing process without addition of amendments.

The pots were then placed in the glasshouse for 2 weeks to allow the soil and amendments time to equilibrate before planting 12-week old in vitro bananas (*Musa* AAA, Cavendish subgroup, cv. Williams) into each pot. Half of the soil was removed from each pot to allow sufficient room to plant the in vitro plantlets and backfilled with the remaining soil. Pots were tapped several times to achieve a uniform bulk density. The banana plants were allowed to grow in the amended soil for a further 3 days and then inoculated with 860 motile *R. similis*, taken from carrot cultures (Moody et al. 1973).

Design and management

All treatments comprising of soil type and amendment combinations were replicated four times in a randomised block design. Pots were maintained in the glasshouse at 20–30°C and received 5 mm of water daily through an automated sprinkler system. Fertiliser was applied (5 g Osmocote Plus Mini™ 16:8:11 N:P:K plus trace elements) at planting and additional soluble fertiliser (Thrive™ 27:5.5:9 N:P:K) was applied 4 and 8 weeks after bananas were planted in pots. The plants were harvested 12 weeks after inoculation with *R. similis*, with plant growth and nematode parameters determined.

Plant growth

Plant height and number of leaves were determined when plants were inoculated with *R. similis* and at four weekly intervals until the termination of the experiment after 12 weeks (84 days). The relative change in plant height and leaf number for each treatment was calculated for each 4 week period. The change in height or leaf number was divided by the number of weeks between assessments, to obtain an average weekly change in plant height and leaf emergence.

Plant height and leaf number were determined at the termination of the experiment, as well as, the area of the last fully emerged leaf, plant dry weight and fresh root weight. The area of the last fully emerged leaf was estimated by determining the length and width of the leaf at its widest point and multiplying by 0.83 (Turner 1972). Plant dry weight was determined by placing shoots of banana plants in an oven at 75°C for 5 days. Banana fresh root weight was determined by washing the soil from the roots and allowing the roots to air dry for 30 min before weighing. After determining the fresh root weight nematodes were extracted from the banana roots.

Nematode extraction

R. similis was extracted from the roots by cutting the roots into 1–2 cm pieces and placing them on a coarse screen in a misting cabinet for 7 days (Hooper 1986). *R. similis*, washed from the roots, were collected at the base of the container with excess misting water. Nematodes were captured by passing the water through a 25 μm sieve. The nematodes were backwashed from the sieve and collected in a 30 mL vial for counting.

Soil nematodes were extracted by placing 200 g of soil collected from pots on a single layer of tissue, contained within a mesh basket (Whitehead and Hemming 1965). The basket was placed in 200 mL of water within a tray and

Table 2 Amendments applied to soil in which bananas were grown, their rate of application (air dried) and chemical composition (air dried), including heavy metal composition of biosolid, compost, mill mud and mill ash

Amendment	Rate t ha^{-1}	C%	N%	C:N ratio	P%	K%	Ca%	Mg%	S%	Na%	Si%	Cu mg kg^{-1}	Zn mg kg^{-1}	Mn mg kg^{-1}	Fe mg kg^{-1}	Al mg kg^{-1}	B mg kg^{-1}
Banana residue	40	46.8	1.8	25.7	0.35	2.0	1.5	0.72	0.10	0.4	2.3	59.3	110	1300	24000	55000	<0.3
Biosolid	40	7.1	0.8	8.7	0.39	1.5	1.0	0.77	0.64	2.24	1.4	14.9	95	180	330	90	54
$CaSiO_3$	5	–	–	–	–	–	34.5	–	–	–	24.2	–	–	–	–	–	–
Grass hay	40	44.7	2.3	19.6	0.29	3.2	0.5	0.16	0.54	0.76	1.3	9	22	190	170	140	6
Legume hay	40	45.8	3.2	14.5	0.25	1.7	1.7	0.52	0.35	0.30	0.6	6.1	15	61	880	700	45
Mill ash	40 & 120	19.4	0.3	64.7	0.27	0.3	0.7	0.16	0.05	0.01	25.3	32.4	76	930	13000	38000	<0.3
Mill mud	40	19.8	1.4	13.7	0.39	0.5	6.3	0.23	0.40	0.52	15.7	247.8	460	270	13000	11000	<0.3
Molasses	300 L ha^{-1}	–	0.5	–	0.06	3.8	0.7	0.25	0.43	0.04	–	2	8.8	70	350	110	3
MW Compost	40	29.0	1.8	15.7	0.28	0.1	32.0	0.19	0.13	0.05	15.9	56.8	120	83	670	2200	11
	Cd mg kg^{-1}	Pb mg kg^{-1}	Co mg kg^{-1}	Cr mg kg^{-1}	Cu mg kg^{-1}	Ni mg kg^{-1}	Zn mg kg^{-1}										
Biosolid	0.48	9.2	1.1	7.6	62.2	5.4	139										
Mill ash	0.20	15.7	10.8	55.6	74	27.4	140										
Mill mud	0.17	14.9	14	74.4	76.1	48.2	143										
MW Compost	2.12	780	17	117	293	84.9	785										

maintained at 25°C. After 48 h, nematodes contained within the water of the tray were collected on a 25 μm sieve. The nematodes were backwashed from the sieve and collected in a 30 mL vial. The total number of nematodes extracted from 200 g of soil was determined. Using a compound microscope, nematodes were identified to genera for plant-parasitic nematodes or families for non-parasitic nematodes and assigned to trophic groups according to Yeates et al. (1993) and indices calculated. From each sample all or 100 individual nematodes were identified based on stoma shape and morphological characteristics at 400x magnification (Goodey 1963).

Nematode indices

Indices of the nematode community composition were calculated from the number of nematode taxa extracted from the soil of each pot. Nematode diversity was determined using the Shannon-Weiner index, $H' = -\Sigma p_i \log_e p_i$ and dominance calculated using Simpson's index of dominance $\lambda = \Sigma (p_i)^2$, where p_i is the proportion of individuals in the i^{th} taxon (Yeates and Bongers 1999).

Additionally, the weighted faunal analysis concept was applied, without plant-parasites, to determine the basal, structure and enrichment conditions of the soil food web (Ferris et al. 2001). The enrichment index (EI) assesses the resources available to the soil food web and response by primary decomposers to those resources. The structure index (SI) is a measure of the number of trophic layers in the soil food web and the potential for regulation by predators. These indices were calculated as $EI = 100[e/(e+b)]$ and $SI = 100[s/(s+b)]$, where e, s and b are the abundance of nematodes in guilds representing enrichment (e) [B1 and F2, where B = bacterivores, F = fungivores, and numbers represent the coloniser–persister (c-p) value 1–5 (Bongers 1990)], structure (s) (B3–B5, F3–F5, O3–O5, P2–P5, where O = omnivores, P = predators) and basal (b) (B2 and F2) nematode communities (Ferris et al. 2001). The channel index (CI), is an indication of the decomposition channel of nutrients calculated as $CI = 100[0.8\ F2/(3.2B1 + 0.8\ F2]$ (Ferris et al. 2001). A low value suggests a primarily bacterial decomposer community whereas a high value indicates a fungal-dominated decomposer nematode community (Hohberg 2003).

Statistics

Significant differences were determined using a two-way ANOVA for treatment and soil effects. If statistical differences were found, the means were separated using the LSD method using Genstat 11 statistics package (VSN International). Total nematode numbers and number of nematodes in each trophic group per 100 g of soil were transformed using $\ln(x+1)$, prior to analyses, to comply with assumptions of normally distributed data. The major nutrient (N, P, K, Ca, Mg and C) content of amendments were subjected to principal component analysis (PCA) and a biplot was constructed using the correlation matrix method. The $CaSiO_3$ was omitted from the PCA of the amendments as it did not contain all of the major nutrients listed above. The soil nematode community data was subjected to PCA. However, the PCA did not add to the findings determined using ANOVA and therefore the results were not included. All statistical analysis was performed using Genstat 11 (VSN International).

Results

Amendments composition

The banana residue, grass hay and legume hay all had carbon contents greater than 40%, suggesting that at a rate of 40 t ha^{-1}, greater than 16 t ha^{-1} of carbon was mixed in with the soil (Table 2). Conversely, biosolid applied at 40 t ha^{-1} contained 7% carbon, so that only approximately 3 t ha^{-1} of carbon was mixed with the soil (Table 2). Biosolids also had the lowest C:N ratio (8.7) of the amendments, where as mill ash had the highest C:N ratio (64.7) (Table 2). Legume hay had the highest nitrogen content (3.2%) of the amendments, which meant that at a rate of application of 40 t ha^{-1}, 1 280 kg N ha^{-1} was potentially available if all nitrogen in the legume hay was mineralised. The grass hay, banana residue and MW compost also contained relatively high amounts of N and could potentially supply more than 700 kg N ha^{-1} if all N was mineralised. The MW compost, sourced from a municipal supplier, contained 32% Ca, five times more than the next highest amendment, mill mud, which contained 6.3% Ca (Table 2). The MW compost was also found to have high amounts of lead, 780 mg kg^{-1}, which is above an acceptable level and 50 times higher than any other amendment. The mill mud, mill ash and banana residue all contained high levels of metals, such as Fe and Al, relative to other amendments (Table 2).

The first two principal components from the PCA of the macronutrient composition of the amendments (C, N, P, K, Mg and Ca) explained over 63% (PC1=35.3 and PC2=28.0) of the variation of nutrient composition of the amendments (Fig. 1). The PCA grouped legume hay, grass hay and banana trash together. The N, C and Mg vectors of the biplot appeared to be positively associated with the grouping of legume hay, grass hay and banana residue (Fig. 1). Conversely, mill ash was on the opposite side of the biplot from the origin relative to legume hay, grass hay and banana trash (Fig. 1). The N and C content of the amendments gave the two highest latent vector loadings in the first principal component (−0.52 and −0.51 respectively), which suggested that these two nutrients were able to explain most of the variation in nutrient content of the amendments.

R. similis suppression

There was no significant interaction between soil type and the amendment's ability to suppress *R. similis* in the roots of the banana plants. Therefore, the results of *R. similis* recovery from the three different soils used in the experiment were pooled across soil types (Fig. 2). The amendments legume hay, grass hay, banana residue and mill-mud significantly reduced ($P<0.05$) the number of *R. similis* in the roots of banana plants relative to the untreated control (Fig. 2). The legume hay was significantly better than all other treatments, with less than 6% of the number of *R. similis* relative to the untreated plants. The grass hay, banana residue and mill mud treatments had a 5-fold reduction in the number of *R. similis* recovered from the roots of bananas compared to the untreated plants (Fig. 2). The $CaSiO_3$, molasses, MW compost, mill ash and biosolid treatments all had statistically similar numbers of *R. similis* in the roots of bananas relative to the untreated control (Fig. 2). Other plant-parasitic nematode species, *Helicotylenchus dihystera*, *Rotylenchulus reniformis* and *Meloidogyne* spp., were extracted in low numbers from the roots of

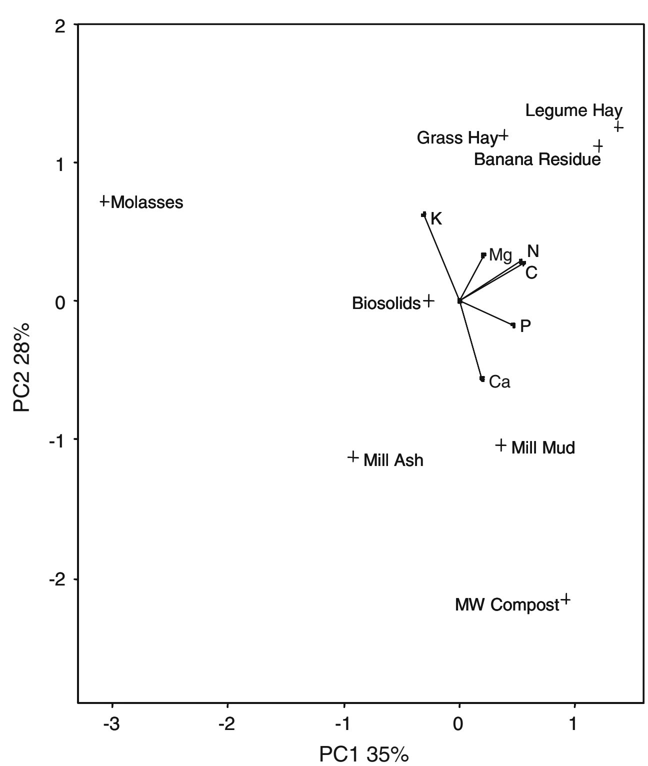


Fig. 1 Principal component biplot for the characterisation of soil amendments used in a pot experiment for suppression of *R. similis* in bananas based on macronutrient content of the amendments

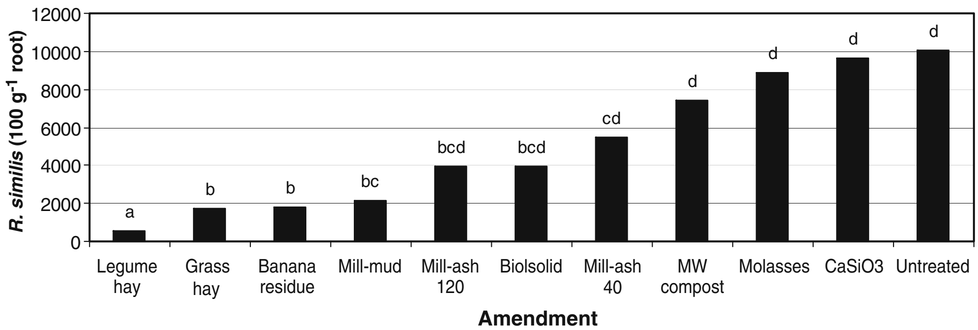


Fig. 2 Means of *R. similis* recovered from the roots of banana plants after 12-weeks from being inoculated, grown in soil with different amendments and pooled across three soil types. (Bars, with the same letter above are not significantly different from one another at P=0.05)

the banana plants, but gave inconsistent recovery, and therefore the data was not included.

Plant growth

The application of biosolid to the soil significantly reduced the growth of plants relative to the untreated soil at 28 days (Table 3). The plants grown in the biosolid amended soil were approximately two-thirds of the shoot dry weight and root fresh weight of the untreated plants and exhibited an 18% decrease in the size of the last fully emerged leaf (Table 3). In the first 4 weeks after planting the bananas, the biosolid treatment resulted in a four-fold reduction in plant height and significantly reduced leaf emergence relative to the untreated soil (Table 3). The height and leaf emergence of the plants grown in the biosolid treated soil were not significantly different from the untreated plants in the mid 4-week period (28 to 56 days) and were significantly better in the final 4-week period of the experiment (56 to 84 days) (Table 3).

All treatments, except the mill mud, mill ash applied at 120 t ha^{-1} and $CaSiO_3$, significantly slowed the growth of banana plants in the first 4-week period (Table 3). In the second 4-week period, between 28 and 56 days after commencing the experiment, grass hay, legume hay and banana residue all significantly increased the growth of the banana plants relative to the untreated plants, but there was no difference in the emergence of leaves over the same period (Table 3). In the final 4-week period of the experiment, between 56 and 84 days, grass hay, legume hay and banana residue as well as the biosolid treatments had significantly greater plant height increases and leaf emergence rates relative to the untreated soil (Table 3).

At the termination of the experiment, banana plants grown in mill mud amended soil had developed significantly greater shoot and root weights, having 1.4 times greater dry shoot weight and fresh root weight than the untreated plants (Table 3). Banana plants grown with additional banana residue had significantly greater shoot dry weight than the untreated soil (Table 3). The incorporation of banana residue also produced plants with the greatest area of the last fully emerged leaf, but this was not significantly greater than the untreated plants (Table 3). Furthermore, the application of mill ash at 120 t ha^{-1} produced significantly greater fresh root weight than the untreated soil (Table 3).

Table 3 Growth of banana plants with different amendments applied to soil in a pot experiment

Amendment	Rate (t ha^{-1})	Increase in plant height (mm week^{-1})			Leaf emergence (leaves week^{-1})			Last fully emerged leaf (cm^2)	Shoot dry weight(g)	Root fresh weight (g)
		28 days	56 days	84 days	28 days	56 days	84 days			
Untreated	0	36f	22a	7a	1.1bcd	0.7n.s.	0.4ab	43.6ab	17.8bcd	53.8bc
Banana residue	40	27cd	37c	14bc	1.1abc	0.8n.s.	0.6de	47.7a	24.5e	66.9cde
Biosolid	40	8a	24ab	13bc	1.0a	0.7n.s.	0.6de	35.8c	11.6a	36.0a
$CaSiO_3$	5	31def	25ab	6a	1.1bcd	0.7n.s.	0.4abc	40.6bc	16.8bc	56.4bcd
Grass hay	40	24bc	39c	16cd	1.1bc	0.8n.s.	0.6e	46.8a	21.2cde	61.4bcde
Legume hay	40	21b	29bc	19d	1.0ab	0.8n.s.	0.6de	40.6bc	15.7ab	50.5abc
Mill ash	120	33ef	26ab	9ab	1.2de	0.7n.s.	0.5bcd	42.8ab	22.0de	71.4de
Mill ash	40	29cde	26ab	8a	1.1cde	0.7n.s.	0.5cd	40.6bc	21.1cde	52.9abc
Mill mud	40	36f	28ab	8a	1.2e	0.8n.s.	0.4abc	42.5ab	25.9e	75.9e
Molasses	300 L ha^{-1}	24bc	28ab	7a	1.1abc	0.8n.s.	0.5cd	38.6bc	16.6bc	46.1ab
MW Compost	40	27cde	25ab	6a	1.1cde	0.8n.s.	0.4a	41.5b	18.1bcd	54.8bcd

Means in columns followed by the same subscript are not significantly different from one another at P=0.05

Banana plants grown in the Innisfail soil series had a significantly greater increase in plant height in the first 28 days relative to the Mundoo and Coom soil series (Table 4). In the final 28 days of the experiment, plants grown in the Coom soil series had a significantly reduced leaf emergence rate relative to the Mundoo soil series. Similarly at the conclusion of the experiment the plants grown in the Coom soil series had at significantly reduced last fully emerged leaf area and root weight, relative to the other two soils used in the experiment (Table 4).

Amendment effects on soil nematode community structure

There was a significant interaction between the application of some of the amendments across the three different soils used in the experiment in terms of number of soil nematodes recovered from different nematode trophic groups (Fig. 3).

Plant-parasitic nematodes extracted from the soil included *R. similis*, *H. dihystera*, *R. reniformis* and *Meloidogyne* spp. The application of banana residue, legume hay, grass hay and biosolid amendments were able to reduce the total number of plant-parasitic nematodes across all genera in all three soils relative to the untreated soil (Fig. 3a). However, some amendments were more effective at reducing the population of plant-parasitic nematodes in particular soil types. For example, the application of mill mud reduced the number of plant-parasitic nematodes in the Coom and Innisfail soils, but did not in the Mundoo soil (Fig. 3a). Conversely, $CaSiO_3$ and molasses resulted in no reduction of plant-parasitic nematode numbers in any of the soils relative to the untreated soil (Fig. 3a).

Bacterivores extracted from soil samples were identified and belonged primarily to the nematode families Cephelobidae, Rhabditidae, Pangrolaimidae, Wilsonematidae and Prismatolaimidae. The application of grass hay, MW compost and mill mud to the Mundoo soil increased the number of bacterivores, relative to the untreated, but the amendments had little effect on bacterivore numbers in the Innisfail and Coom soils (Fig. 3b). There were relatively high numbers of bacterial feeding nematodes in the untreated Coom soil and the addition of amendments did not significantly alter the number of bacterivores recovered from the soil relative to the control (Fig. 3b). However, the application of the amendments to the Coom soil caused both an increase and decrease in numbers of bacterivores, with the biosolid treatment having the highest recovery of bacterivores and significantly greater numbers of bacterivores than the mill mud treated soil (Fig. 3b). Similarly, the legume hay applied to the Innisfail soil series had a significantly greater number of bacterivores recovered from soil relative to the mill mud treated Innisfail soil (Fig. 3b).

Fungivores recovered from soil extracts were identified as belonging to the families Aphenchidae or Tylechidae. The application of mill mud significantly reduced the number of fungivorous nematodes in the Innisfail soil relative to the untreated soil (Fig. 3c). The mill mud treatment had the lowest number of fungivorous nematodes in the Coom and Innisfail soils. There were significantly fewer fungivores in the mill mud treated Coom soil relative to the legume hay treatment (Fig. 3c). Similarly, there were fewer fungivores in the mill mud treated Innisfail soils relative to mill ash 40 t ha^{-1}, MW compost, grass hay, legume hay and untreated soil (Fig. 3c). The application of grass hay, MW compost, banana residue and mill ash at 120 t ha^{-1}, to the Mundoo soil significantly increased the number of fungivores in the soil relative to the application of $CaSiO_3$, but not the untreated soil (Fig. 3c).

All omnivores identified belonged to the family Dorylaimidae. Very low numbers of omnivorous nematodes were extracted from the untreated Mundoo soil and only the application of grass hay significantly increased the number of omnivores (Fig. 3d). Furthermore, the application of grass hay to the Coom soil significantly increased the number omnivorous nematodes relative to the untreated soil (Fig. 3d). Conversely, the application of biosolid and $CaSiO_3$ significantly reduced the number of omnivores in the Coom soil relative to the untreated soil (Fig. 3d). Four amendments, mill ash 40 t ha^{-1}, MW compost, grass hay and banana residue applied to the Innisfail soil, all significantly increased the number of omnivorous nematodes recovered relative to the untreated Innisfail soil (Fig. 3d).

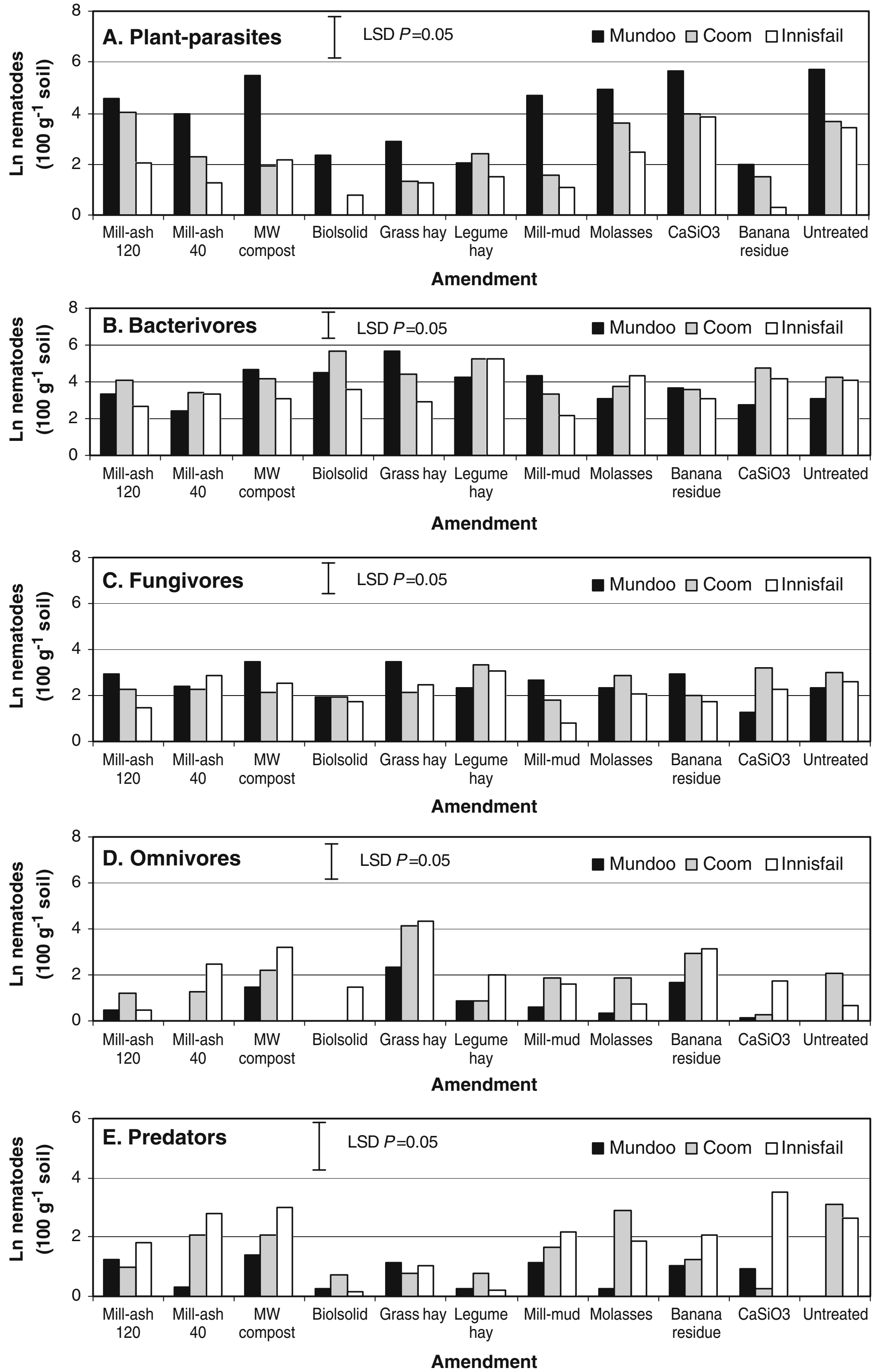
Predatory nematodes extracted from soil samples belonged to the families Mononchidae or Tripylidae. There were

Table 4 Growth of banana plants in a pot experiment using soils common in banana production in north Queensland

Soil	Increase in plant height (mm $week^{-1}$)			Leaf emergence (leaves $week^{-1}$)			Last fully emerged leaf (cm^2)	Shoot dry weight (g)	Root fresh weight (g)
	28 days	56 days	84 days	28 days	56 days	84 days			
Coom	26a	26n.s.	9n.s.	1.1n.s.	0.8n.s.	0.4a	39.5b	17.2a	43.5a
Innisfail	29b	28n.s.	9n.s.	1.1n.s.	0.8n.s.	0.5ab	43.0a	20.3b	54.9b
Mundoo	26a	30n.s.	11n.s.	1.1n.s.	0.8n.s.	0.5b	42.8a	19.6ab	69.5c

Means in columns followed by the same subscript are not significantly different from one another at $P=0.05$

Fig. 3 Effects of amendments applied to three different soil types on the trophic groups of soil nematodes (Bars represents LSD values at P=0.05)



significantly more predatory nematodes in the untreated Coom and Innisfail soils relative to the untreated Mundoo soil (Fig. 3e). No amendment applied to the any of the three soils significantly increased the number of predatory nematodes relative to the corresponding untreated soil (Fig. 3e). However, the number of predatory nematodes was significantly reduced in Coom and Innisfail soils following the application of biosolid, grass hay and legume hay relative to the untreated soil (Fig. 3e). Furthermore, the application of mill ash 120 t ha^{-1} and $CaSiO_3$ to the Coom soil significantly reduced the number of predatory nematodes relative to the untreated Coom soil (Fig. 3e).

Unlike the number of nematodes in each trophic group, there was no significant interaction between amendments and soil type in relation to nematode community indices and therefore, the data for the different indices have been pooled for amendment effects across soil types (Table 5). The application of biosolids significantly decreased the number of taxa, diversity index and increased the dominance index of soil nematode community relative to the untreated soil (Table 5). Conversely, the application of banana residue was the only treatment to significantly increase the diversity index of nematodes in the soil compared to the untreated soil (Table 5). Furthermore, mill mud was the only treatment to significantly reduce the dominance index of nematode taxa in the soil compared to the untreated soil (Table 5). The application of the MW compost significantly increased the number of nematode taxa relative to all other treatments (Table 5).

Mill mud, banana residue, MW compost and grass hay treatments all significantly increased the structure index relative to the untreated soil (Table 5). Conversely, the application of biosolids significantly reduced the structure index and increased the enrichment index compared to the untreated soil (Table 5). Furthermore, banana residue, MW compost and legume hay significantly increased the enrichment index relative to the untreated soil (Table 5). The application of biosolid and legume hay exhibited a predominantly bacterial decomposition pathway of nutrients having a significantly lower channel index relative to the untreated soil (Table 5). Seven treatments, banana residue, biosolid, MW compost, grass hay, legume hay, mill ash 40 t ha^{-1} and mill mud all significantly reduced the proportion of plant-parasitic nematodes recovered from the soil relative to untreated soil (Table 5). The mill ash 120 t ha^{-1}, molasses and $CaSiO_3$ treated soils all had statistically similar proportion of plant-parasitic nematodes in the soil community compared to the untreated soil (Table 5).

Discussion

The application of amendments to soil was found to affect different soil properties depending on the type of amendment applied. In addition, amendments that had high organic carbon contents induced high levels of suppression of the population of *R. similis* in the roots of banana plants. In particular, legume hay, grass hay, banana residue and mill mud treatments were associated with significant reductions in the number of *R. similis* in the roots of banana plants relative to the untreated plants. These four treatments also had the lowest proportion of plant-parasitic nematodes in the soil, relative to the untreated soil. The suppressive mechanism, which reduced the number of plant-parasitic nematodes in the soil, was unknown, so it was categorised as general suppression. The application of amendments that were high in organic carbon altered many biological properties in the soil, but there are other possible explanations for the observed reduction of *R. similis* in the roots of bananas. The number of *R. similis* that penetrated the roots of bananas following inoculation may have been reduced due to the production of metabolites resulting from the decomposition of amendments with high organic carbon contents. It is also possible that these amendments stimulated bacteria and fungi, which were antagonistic to nematodes preventing them from penetrating the roots or reducing their multiplication once they had penetrated banana roots. The application of grass hay and banana

Table 5 Effects of soil amendments on soil nematode indices

Amendment	Rate (t ha^{-1})	Number of taxa	Diversity (H′)	Dominance (λ)	Structure index	Enrichment index	Channel index	Plant-parasitic nematodes (%)
Untreated	0	7.8d	1.48bc	0.49bcd	47b	67def	20abc	48a
Banana residue	40	7.9d	1.70a	0.43cde	78a	81b	10cde	9e
Biosolid	40	5.2a	0.93e	0.62a	33c	96a	1e	6e
$CaSiO_3$	5	7.1bc	1.24d	0.55ab	42bc	71cdef	17bc	50a
Grass hay	40	6.8b	1.32cd	0.52bc	80a	75bcde	22ab	3e
Legume hay	40	6.6b	1.32cd	0.52bc	55b	93a	5de	6e
Mill ash	120	6.5b	1.40cd	0.52bc	46bc	65f	31a	47a
Mill ash	40	7.8d	1.67ab	0.49bcd	51b	67ef	29a	33bc
Mill mud	40	7.8d	1.69ab	0.37e	72a	67ef	16bc	24d
Molasses	300 L ha^{-1}	7.7cd	1.46c	0.50bc	52b	77bcd	14bcd	42ab
MW Compost	40	8.8e	1.67ab	0.41de	71a	79bc	11bcde	28cd

Means in columns followed by the same subscript are not significantly different from one another at $P=0.05$

residue caused an increase in the number of omnivorous nematodes, so predation by other nematodes may also have had a role in the suppression of *R. similis*.

Stirling et al. (2005), similarly found suppression of plant-parasitic nematodes in sugar cane soil amended with high amounts of organic carbon. This research was not able to define the suppressive mechanism causing the reduction of plant-parasitic nematodes, but they postulated that there was an overall shift in the soil food web causing a cascade of changes in biological properties in the soil that could lead to nematode suppression. A change in biological properties in amended soils was also evident in the current investigation, with significantly different numbers of nematodes belonging to different trophic groups, found in soils with different amendments. Similarly, the nematode community indices were also significantly different with the different amendment treatments, with amendments high in C increasing the structure index. Therefore, applications of amendments high in carbon appear to be able to induce suppression of plant-parasitic nematodes in the soil and roots of banana plants through changes to the biological composition of the soil food web.

From this experiment it appeared that grass hay, legume hay and banana residue, with high carbon content, increased the number of beneficial organisms in the soil. The amendments with a relatively high N and C contents gave the greatest suppression of *R. similis*. Furthermore, these amendments could be distinguished from others by use of the PCA based on their macronutrient content (Fig. 1). Therefore, the nutrient content of amendments, in particular their N and C contents, could be regarded as indicators of the suppressive potential of the amendment. Further investigation is required to establish a range of amendments with differing N and C contents to determine their suppression of plant-parasitic nematodes and their impact on the soil nematode community.

The biosolid had the lowest C:N ratio, but was able to significantly reduce the number of plant-parasitic nematodes in the soil. However, the addition of biosolid was phytotoxic to banana plants, causing a reduction in plant growth in the first 8-weeks of the experiment. The application of biosolids also caused significant changes in nematode community composition with an increase in the number of bacterivores, a decline in the number of predators and omnivores and reduction in the diversity and structure indices of the nematode community. This suggested that the reduction in plant-parasitic nematode populations in the soil caused by biosolids may have been due to chemical effects from the decomposition of the biosolid that were toxic to most soil organisms as well as the banana plant. Therefore, the suppression of plant-parasitic nematodes in the roots of bananas following biosolid application was not due to increased nematode antagonistic organisms, but possibly from induced chemical changes in the soil. Increased bacterial activity was suggested by Chavarria-Carvajal et al. (2001) and Lazarovits et al. (2003) to favour plant-parasitic nematode suppression. However, the increase in bacterial feeding nematodes, mediated through the application of biosolids, was not the best method to develop suppression of *R. similis* in bananas. Therefore, some knowledge of how amendments affect soil biology and how their degradation impacts on soil biology is important in order to develop strategies to suppress plant-parasitic nematodes in agricultural crops.

Early growth of the banana plants was slowed by most amendments, except mill mud, mill ash and $CaSiO_3$, relative to plants grown in untreated soil. This is possibly because of the nutrient release rate from the amendments, with nutrient draw-down reducing nutrient availability for early plant growth for amendments with high C:N ratios. Even though the amendments were allowed to equilibrate for 2 weeks prior to planting bananas, this may not have been sufficient time to prevent nutrients being immobilised during the initial decomposition of the amendments. However, after 28 days, any effects of nutrient draw-down were overcome and amendments with high carbon contents, banana residue, grass hay and legume hay, produced significantly taller plants than the untreated control. The size of the last fully emerged leaf indicated how the plants were growing at the termination of the experiment. Only the biosolid treatment had a significantly smaller leaf area than the untreated soils. This suggested that nutrient draw-down effects of amendments on early plant growth had been overcome at the conclusion of the experiment, 12 weeks after planting. Furthermore, plant growth appeared to have been stimulated by the use of mill mud producing significantly greater shoot and root dry weight, as well as promoting nematode suppression. Therefore, the application of amendments could act as a slow release source of nutrients to sustain banana growth. This is particularly important when bananas are grown in the wet tropics, as nutrient leaching due to heavy rainfall can be a serious problem (Kleiese et al. 1997; Moody and Aitken 1997).

The soil type used in the experiment was an important variable in determining the growth of the plant and structure of the nematode community following the addition of amendments. Better plant growth was observed in the Innisfail soil series in the first 4 weeks of the experiment. This could have been due to a higher initial nutrient content relative to the other two soils. At the termination of the experiment poor plant growth was measured in the Coom soil relative to the Innisfail and Mundoo soil types. The poor structure of the Coom soil appears to have been the factor contributing to the reduction in plant growth at the termination of the experiment. There were no significant interactions between amendments and soil type affecting plant growth, which suggested that the rate and types of amendments, or the time period for the experiment were insufficient to improve the structure of the Coom soil for the growth of bananas.

The different soils also had a different inherent nematode community structure. The application of amendments to the soil changed the nematode trophic group structure, as they responded differently in the different soils. The application of molasses to the Coom soil had little affect on the nematode community composition relative to the untreated soil. Relative to the untreated Mundoo soil, $CaSiO_3$, molasses and mill ash (40 t ha^{-1}) all had similar nematode community structures. However, grass hay and banana residue tended to significantly increase the number of omnivores across all soil types. These results emphasise that the application of amendments to different soils may not produce the same result in terms of nematode suppression because of the inherent soil biological properties that existed before the amendments were applied. Therefore, there needs to be a better understanding of the soil conditions that favour the increase in plant-parasitic nematode antagonistic organisms and how they may be stimulated by the addition of amendments. The change in biological factors in the soil over time as decomposition of organic amendments progresses is also needed. Changes in trophic groups of nematodes may be used as indicators of shifts in the types of antagonistic organisms active in the soil at a specific point in time. For example, as decomposition of amendments occurs bacteria may be active at first, followed by fungal antagonists and lastly by predators.

Further work is required to determine efficacy of amendments in field conditions and to determine how amendments high in C, such as grass hay, can be applied more efficiently and reliably. Furthermore, investigations are required that allow the use of amendments to be incorporated with current agronomic practices in the production of bananas and integrated with other nematode management practices. For example, the use of grassed fallows that are resistant to *R. similis* followed by the incorporation of the organic material may give greater, durable suppression of plant-parasitic nematodes in commercial banana production systems.

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