

ORGANIC AMENDMENTS ENHANCE BIOLOGICAL SUPPRESSION OF PLANT-PARASITIC NEMATODES IN SUGARCANE SOILS

By

G.R. STIRLING^{1,2}, E.J. WILSON^{1,2}, A.M. STIRLING², C.E. PANKHURST^{1,3},
P.W. MOODY^{1,4} and M.J. BELL^{1,5}

¹*Sugar Yield Decline Joint Venture*

²*Biological Crop Protection Pty Ltd., Brisbane*

³*CSIRO Land and Water, Townsville*

⁴*Department of Natural Resources and Mines, Indooroopilly*

⁵*Agency for Food and Fibre Science, QDPI, Kingaroy*

biolcrop@powerup.com.au

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Abstract

Previous research has shown that population densities of plant-parasitic nematodes are reduced when a legume crop is grown in rotation with sugarcane. However, this effect is only temporary, as nematodes usually return to high densities within 12 months of planting sugarcane. This rapid resurgence suggests that natural enemies that normally keep plant-parasitic nematodes under control in natural environments may be depleted by the soil-management practices used to grow sugarcane. This paper describes an experiment in which organic materials were added to sugarcane soils in an attempt to enhance biological activity and increase the suppressiveness of soils to plant-parasitic nematodes. The amendments used were sawdust, sugarcane trash, grass hay and legume hay with or without nitrogen, and feedlot manure, poultry manure, chitin and mill mud without additional nitrogen. The chemical and biological changes occurring during the decomposition process were monitored for 12 months, while the capacity of amended soils to suppress lesion and root-knot nematodes was assessed periodically using bioassays. Seven months after amendments were incorporated, soils amended with sawdust, sugarcane trash or grass hay were more suppressive to root-knot nematode than soils amended with nitrogenous materials. Sugarcane grown in soil amended 6 months previously with sawdust, sugarcane trash, grass hay or lucerne hay had 78%, 61%, 96% and 92%, respectively, fewer lesion nematodes in roots than sugarcane growing in non-amended soil. Low concentrations of nitrate nitrogen in the soil, a fungal dominant soil biology and high numbers of omnivorous nematodes were most closely associated with suppression. These results indicate that the biology of sugarcane soils can be altered by changing the quality and quantity of organic inputs. Amendments with high C/N ratios are most effective in enhancing biological control activity against plant-parasitic nematodes.

Introduction

Some nematodes feed on plants and are important pests, but the majority of species are beneficial, feeding on bacteria, fungi, algae, nematodes and other microfauna (Yeates, 1999). Pest species are the dominant group of nematodes in Queensland sugarcane fields. Plant-parasites comprised 33, 36, 30 and 40% of the nematode community following a short fallow at Tully, Ingham, Mackay and Bundaberg, respectively, and multiplied to 38, 57, 68 and 67%, respectively, of the population within 11–16 months of planting sugarcane (Stirling *et al.*, 2001). The most widely distributed species are lesion nematode (*Pratylenchus* spp.), root-knot nematode (*Meloidogyne* spp.) stunt nematode (*Tylenchorhynchus* spp.), stubby root nematode (*Paratrichodorus* spp.), reniform nematode (*Rotylenchulus* spp.), spiral nematode (*Helicotylenchus* spp.), ring nematode (*Criconemella* spp.), and dagger nematode (*Xiphinema* spp.). At least five of these are found in most fields (Blair *et al.*, 1999a,b). This pest complex produces relatively non-specific symptoms, but lack of fine roots, swelling of root tips, stunting of lateral roots, blackening and discoloration of fine roots, and occurrence of pinkish-purple lesions on primary roots are indicative of nematode damage (Spaull and Cadet, 1990).

Currently, nematode control in sugarcane is based on organophosphate and carbamate nematicides. They are some of the most toxic agricultural chemicals in use today, are relatively expensive, control nematodes for no more than about 3 months, affect both pest and beneficial nematode species and can contaminate groundwater (Bunt, 1987; Thomason, 1987). Sugarcane varieties with some degree of resistance to root-knot nematode have been bred, but it is unlikely that varieties with resistance or tolerance to the diverse range of nematodes that attack sugarcane will ever be developed (Spaull and Cadet, 1990). Thus, cultural and biological controls must form the basis of future nematode management strategies.

The introduction of a rotation crop into the sugarcane farming system is the first step towards redressing the imbalance between pest and beneficial components of the nematode community. A single crop of soybean, for example, reduces populations of lesion nematode at planting by 44–89% and increases the ratio of beneficial to plant-parasitic species from 2:1 to 20:1 compared to fields where sugarcane is grown continuously (Stirling *et al.*, 2002). However, the effect of a rotation crop is only temporary, as pest species return to high densities within 12 months of planting sugarcane (Stirling *et al.*, 2002). This resurgence is partly due to the availability of sugarcane roots as a food source. However, it is also possible that the suppressive agents that normally keep plant-parasitic nematodes under control in natural environments may have been depleted by the soil management practices used in the sugar industry.

Two ways of changing the soil biology to enhance suppression to plant-parasitic nematodes are to increase the level of organic matter inputs into cane-growing soils, or to change the composition of those inputs. When organic matter is added to soil, bacteria and fungi commence the decomposition process and multiply rapidly, while populations of other soil organisms (e.g. protozoans and algae) also increase. Bacterial and fungal-feeding nematodes then multiply as more food becomes available. Finally there is an increase in numbers of omnivorous and predatory nematodes and in the activity of predatory fungi such as the nematode-trapping fungi. This results in a more complex food web with enhanced biological control activity (Stirling, 1991).

Here, we describe an experiment to determine whether the addition of organic matter to sugarcane soils enhances biological activity and increases suppressiveness to plant-parasitic

nematodes. We had no idea what materials were likely to be effective, so we assessed a range of organic materials that are readily available within the sugar industry. Chemical and biological changes were monitored for 12 months after the organic materials were added, and bioassays were used to assess suppressiveness to root-knot and lesion nematodes.

Materials and methods

The experiment was established in a sandy loam soil at Bundaberg. The previous sugarcane crop had been harvested 6 months previously and the site prepared by cultivation. In March 2001, sawdust from hardwood species, baled sugarcane trash, grass hay as baled oat chaff, lucerne hay, poultry manure from sheds used for broiler production, feedlot manure from a cattle feedlot, mill mud from a local sugar mill, and chitin as laboratory grade material obtained from Sigma were spread on the soil surface with or without additional ammonium nitrate and incorporated with a rotary hoe to a depth of about 15 cm. Application rates (t dry matter/ha) are listed in Table 1. Carbon inputs were estimated by assuming carbon contents of 40% for plant-derived materials, 25% for feedlot and poultry manure, 47% for chitin, and 30% for mill mud. There were four replicate plots 1.5 m long x 1.5 m wide for all amendments, and eight replicate plots of the non-amended control.

Table 1—Amendments incorporated into soil at Bundaberg in March 2001.

Amendment	Application rate (dry t/ha)	C applied (t/ha)	Additional N (kg/ha)
Nil	0	0	0
Sawdust	50	20	0
Sawdust + N	50	20	200
Sugarcane trash	50	20	0
Sugarcane trash + N	50	20	200
Grass hay	50	20	0
Grass hay + N	50	20	200
Lucerne hay	50	20	0
Lucerne hay + N	50	20	200
Feedlot manure	40	10	0
Poultry manure	40	10	0
Chitin	21	10	0
Mill mud	38	12	0

Soil samples were taken from each plot on 19 April, 19 July and 29 October 2001, and on 21 March 2002 (i.e. 1, 4, 7 and 12 months, respectively, after the amendments were added). At each sampling time, a total of 2.5 L of soil was collected with a spade from five points in each plot at depths of 2–10 cm. Populations of bacteria, fungi and nematodes, measurements of microbial activity and assays for suppression were done on every sample, but a single composite sample obtained by mixing 100 mL of soil from each replicate was used for chemical analyses.

Soil chemistry

Soil pH_w and EC were determined on 1:5 soil water suspensions. Nitrate and ammonium-N were extracted with 2 M KCl and determined by the automatic colorimetric procedure (method 7C2; Rayment and Higginson, 1992). Total soil organic carbon (C) and nitrogen (N) were determined on a Leco C-N combustion analyser.

Soil biology

Populations of bacteria and fungi were estimated by adding 2 g of soil to 18 mL of sterile phosphate-buffered saline (PBS) in a glass bottle and shaking at 250 r/min for 30 minutes. A sample of this suspension (1 mL) was then serially diluted and plated onto 0.1 TSA plus 100 µg/mL cycloheximide (for bacteria) and 0.25 potato dextrose agar plus 100 µg/mL rifampicin and 100 µg/mL novobiocin (for fungi). Microbial biomass C was determined on soil that had been passed through a 2 mm sieve and adjusted to a moisture content of 70% field capacity. A 20 g soil sample was fumigated for 10 days under an atmosphere of CHCl₃ and then extracted with 0.5 M K₂SO₄. Extracts were filtered, digested with K₂Cr₂O₇ and H₂SO₄, and the carbon determined spectrophotometrically at 600 nm using sucrose as a standard (Vance *et al.*, 1987). Microbial activity was estimated by measuring the rate of hydrolysis of fluorescein diacetate (Chen *et al.*, 1988).

Nematodes

Nematodes were extracted by spreading 200 mL of soil on a tray for 4 days (Whitehead and Hemming, 1965) and then concentrating nematodes in the suspension on a 38 µm sieve. Free-living nematodes were separated into four trophic groups and counted: bacterial-feeding nematodes (Rhabditida), fungal-feeding nematodes (Aphelenchina and some Tylenchina), omnivores (Dorylaimida), and predators (Mononchida).

Suppression of root-knot nematode

Two assays were used to measure suppression. In the soil assay, 200 mL samples from each plot were added to polystyrene drinking cups with holes in the base to allow drainage. Four replicate 200 mL cups of pasteurised washed river sand were included as a non-suppressive standard. Eggs of root-knot nematode (*M. javanica*) were obtained by immersing roots of glasshouse-grown, nematode-infested tomato plants in commercial bleach (0.5% available chlorine) for 5 minutes and then washing eggs thoroughly with water on a 38 µm sieve. Cups were inoculated with 4000 eggs and placed in polystyrene boxes covered with moist paper to prevent the soil from drying. After incubation at ambient temperatures for 10 days, nematodes were extracted from soil using the tray method described above and second-stage juveniles of root-knot nematode were counted.

In the plant assay, 400 mL samples of each treatment and the pasteurised sand standard were added to pots, inoculated with 10 000 root-knot nematode eggs and incubated for 10 days as above. A tomato seedling (cv. Tiny Tim) was then planted in each pot and after plants had grown in a glasshouse for about 6 weeks, roots were rated for galling using the 0–10 scale of Zeck (1971). To partially overcome nitrogen deficiency problems in soils amended with materials with a high C/N ratio, plants were watered twice with Aquasol[®], a soluble fertiliser containing 23% N.

To enable comparisons between sampling times, data from the suppression assays are presented as numbers of nematodes in a particular treatment relative to the number in pasteurised sand. Thus, numbers less than 1 indicate suppression, as there were fewer nematodes in the amended soil than in the sand. A low suppression index (e.g. 0.3) indicates a highly suppressive soil, as the number of nematodes in the treated soil is only 30% the number in sand. Data for the root gall assay are presented in a similar way (i.e. the gall rating in amended soil relative to the gall rating in sand).

Suppression of lesion nematode

In September 2001, about 6 months after amendments were incorporated, soil was collected from the four replicate plots of six treatments (nil, sawdust+N, sugarcane trash+N, grass hay+N, lucerne hay+N and mill mud). Each sample was then used to fill two 3 L pots, so that there were eight pots of each treatment. Fertiliser (13%N, 14%P and 13%K) was added at a rate equivalent to 20 g/m² and two single-eye setts of sugarcane (cv. Q188^A) were planted in each pot after setts were dipped in Sportak® (prochloraz) at 0.2 g/L water. Four replicate pots of each treatment were then inoculated with 1300 *P. zae* from carrot cultures, while the other four were not inoculated. Plants were harvested after 68 days and dry weights of tops and roots were measured. Nematodes were extracted from 200 mL soil samples using the tray method (above) and also from sett roots plus shoot roots using a mist extraction technique.

Statistical analyses

Nematode counts were transformed [$\log_{10}(\text{no. nematodes}+1)$] prior to analysis. Data for each sampling time were analysed independently by one-way analysis of variance using Genstat 5 (Lawes Agricultural Trust, Rothamsted Experimental Station, UK). However, because of space constraints only results for the 7 month sampling time are presented.

Comments on temporal effects of treatments are made when particular parameters responded in a different way at other sampling times. The pot experiment on suppression of lesion nematode was analysed as a two-way analysis of variance (amendment x inoculation with *Pratylenchus*), with no blocking.

Results

Soil chemistry

Amendments had major effects on soil chemistry (Table 2). Those with a low C/N ratio (lucerne hay, feedlot manure, poultry manure, chitin and mill mud) increased soil NO₃-N to very high levels, with a consequential reduction in soil pH and increased EC. Lucerne hay also increased concentrations of NH₄⁻N, particularly at the 1- and 4-month samplings (13.1 and 32.2 mg/kg, respectively). The soil C/N ratio in these treatments was relatively low (generally 12–15 at 1 and 4 months, declining to 9–11 at 7 months).

The materials with a high C/N ratio (sawdust, sugarcane trash and grass hay) tended to increase pH, had little impact on EC, and had the highest soil C/N ratios. Levels of NO₃⁻N were very low in the latter treatments at 1 and 4 months, but had increased at 7 months in soils amended with sugarcane trash or grass hay.

Table 2—Effects of organic amendments on various soil chemical parameters 7 months after organic matter was incorporated into soil.

Amendment	pH _w	EC (dS/m)	NO ₃ -N (mg/kg)	C/N
Nil	5.9	0.163	58	13.6
Sawdust	6.5	0.063	1	15.5
Sawdust + N	6.5	0.064	7	14.6
Sugarcane trash	6.2	0.126	17	13.8
Sugarcane trash + N	6.1	0.115	41	13.6
Grass hay	6.0	0.161	42	13.3
Grass hay + N	5.7	0.244	72	13.1
Lucerne hay	5.3	0.693	341	10.3
Lucerne hay + N	5.0	0.884	462	9.8
Feedlot manure	6.0	0.514	93	11.6
Poultry manure	5.3	0.900	474	9.5
Chitin	5.0	0.609	372	9.8
Mill mud	5.5	0.513	309	11.0

Soil biology

One month after application, the organic amendments had impacted on all soil biology measurements, generally increasing microbial biomass, and numbers of bacteria and fungi (data not shown). These increases were generally maintained until 4 months, but by 7 months microbial biomass and bacterial numbers in most amended soils were not significantly different from the non-amended soil (Table 3).

The only exceptions were the sawdust and mill-mud treatments, where microbial biomass was significantly higher than the nil amendment at 7 months. Most amendments increased numbers of fungi at 4 months and these increases were maintained for longer than for bacteria.

At 7 months, amendments with a high C/N ratio had higher numbers of fungi than some of the nitrogenous amendments (Table 3). Microbial activity was initially stimulated by sawdust+N, sugarcane trash+N, grass hay+N, and lucerne hay with or without additional N.

However, there were few differences in microbial activity in amended soils after 4 months and no consistent trends at 7 months (Table 3).

Table 3—Effects of organic amendments on various soil microbial parameters 7 months after organic matter was incorporated into soil.

Amendment	Microbial biomass ($\mu\text{g C/g soil}$)		Microbial activity ($\mu\text{g FDA hydrolysed/h/g soil}$)		Bacteria (Log cfu/g soil)		Fungi (Log cfu/g soil)	
Nil	93	abcd	61	bcd	5.7	b	3.1	ab
Sawdust	125	ef	39	a	5.8	b	3.9	b
Sawdust + N	87	abcd	72	cd	6.1	b	4.0	b
Sugarcane trash	115	def	81	cde	6.1	b	4.2	b
Sugarcane trash + N	110	cde	78	cde	6.0	b	4.0	b
Grass hay	86	abc	79	de	5.8	b	2.7	ab
Grass hay + N	96	abcd	81	de	6.0	b	3.9	b
Lucerne hay	69	a	61	bcd	6.1	b	2.7	ab
Lucerne hay + N	88	abcd	74	cde	6.4	b	4.4	b
Feedlot manure	100	bcde	92	e	3.7	a	1.3	a
Poultry manure	92	abcd	45	ab	6.0	b	4.4	b
Chitin	108	bcde	64	bcd	5.8	b	2.9	ab
Mill mud	144	f	50	ab	5.9	b	1.3	a
Lsd (P = 0.05)	29		21		1.4		2.5	

Means in the same column followed by the same letter are not significantly different (P = 0.05).

Nematodes

All treatments affected either the composition of the nematode community or the population density of various trophic groups and these effects were sometimes maintained for 12 months. At 7 months, for example, there were 4–6 times more free-living nematodes in many amended soils than in the nil treatment (Table 4).

Materials with a high C/N ratio (sawdust, sugarcane trash and grass hay) increased both bacterial and fungal-feeding nematodes. Chitin also increased numbers of both trophic groups, but the other materials with a low C/N ratio (lucerne hay, feedlot manure, poultry manure, chitin and mill mud) had a greater impact on bacterial-feeding species.

Numerically, the omnivores were only a small component of the total population of free-living nematodes. However, they proved to be useful biological indicators because there were clear effects of treatment on their population densities. Lucerne hay, poultry manure and chitin eliminated omnivores, whereas numbers of omnivores increased when materials with a high C/N ratio were added.

Addition of N to amendments with a high C/N ratio generally had little impact on numbers of free-living nematodes at any of the sampling times. Most of the significant effects were observed with bacterial-feeding nematodes, which increased in population density at some sampling times in response to adding N to grass hay or sugarcane trash.

Table 4—Effect of organic amendments on the nematode community and suppression of root-knot nematode (measured as the gall rating or number of second-stage juveniles (J2) relative to pasteurised sand) 7 months after amendments were incorporated into soil.

Amendment	Log (number of nematodes/200 mL soil +1)						Gall rating relative to sand	Number of J2 relative to sand		
	Bacterial-feeders		Fungal-feeders		Omnivores and predators					
Nil	2.88	a	3.03	a	1.19	b	1.08	gh	1.05	ef
Sawdust	3.16	bcde	3.78	e	1.94	cd	0.77	abcde	0.39	abc
Sawdust + N	3.31	cde	3.62	cde	2.18	d	0.85	abcdef	0.45	abcd
Sugarcane trash	3.28	cde	3.66	de	1.98	cd	0.66	a	0.30	ab
Sugarcane trash + N	2.84	ab	3.22	abcd	1.99	cd	0.70	abc	0.19	a
Grass hay	3.12	abcd	3.43	bcde	2.11	d	0.74	abcd	0.37	abc
Grass hay + N	3.47	ef	3.38	abcde	1.98	cd	0.67	ab	0.72	cd
Lucerne hay	3.63	f	3.53	cde	0.40	a	1.16	h	0.78	de
Lucerne hay + N	2.82	a	3.04	ab	0.00	a	0.90	bcdefg	0.56	bcd
Feedlot manure	3.29	cde	3.21	abc	1.25	b	1.01	fgh	0.35	ab
Poultry manure	3.05	abc	3.44	bcde	0.00	a	0.93	cdefgh	0.21	ab
Chitin	3.37	def	3.58	cde	0.00	a	1.00	efgh	0.33	ab
Mill mud	3.37	def	3.38	abcde	1.20	b	0.93	defgh	1.19	f
Lsd (P = 0.05)	0.31		0.43		0.59		0.23		0.36	

Means in the same column followed by the same letter are not significantly different (P = 0.05).

Suppression of root-knot nematode

The suppression indexes for both the soil and plant assays showed that the non-amended soil became less suppressive to root-knot nematode as the experiment proceeded. In the soil assay, the suppression indexes were 0.44, 0.67 and 1.05 at 1, 4 and 7 months, respectively. The corresponding figures for the plant assay were 0.74, 1.04 and 1.08.

Amending soil with organic matter altered the suppressiveness of soil to root-knot nematode, but the effect varied with the type of amendment and with time. Materials with a high C/N ratio were most suppressive, with grass hay being the first to become suppressive, followed by cane trash and then sawdust (data not shown). At 7 months, grass hay, sugarcane trash and sawdust with or without N were suppressive in both plant and soil assays (Table 4). At 12 months, sawdust, sugarcane trash and grass hay with or without additional N were suppressive in the soil assay, but only sawdust, sawdust +N and cane trash were suppressive in the plant assay. The nitrogenous amendments did not consistently increase suppression, but J2 data from the soil assay at 7 months (Table 4) showed that all amended soils except mill mud were significantly more suppressive than the non-amended soil.

Relationships between suppression to root-knot nematode (measured using gall ratings or number of second-stage juveniles relative to sand) and all the chemical and biological parameters

listed in Tables 1–3 were determined using multiple regression analysis for the 4- and 7-month samplings. C/N ratio and populations of omnivore–predators and fungi were positively correlated with suppression, whereas there was a negative relationship with the NO₃ -N concentration (Table 5).

Table 5—Relationships between suppression (measured using gall ratings or number of second-stage juveniles (J2) relative to sand) and chemical and biological parameters 4 and 7 months after incorporating organic amendments into soil.

Sampling time	Measure of suppression	Parameters*	Adjusted R ²	P	
4 months	J2	Omnivore/predators (4)	0.331	0.015	
		C/N (4)	0.254	0.032	
		NO ₃ (4)	0.219	0.045	
		C1 (4)	0.270	0.027	
		C/N (4), Fungi (4)	0.552	0.003	
		Omnivore/predators (4), Fungi (4), NO ₃ (4)	0.581	0.005	
		Galls	C/N (4)	0.554	<0.001
7 months	J2	NO ₃ (4)	0.462	0.003	
		Omnivore/predators	0.318	0.017	
		Fungi (4)	0.620	<0.001	
		Fungi (7)	0.243	0.050	
		Galls	Omnivore/predators (7)	0.472	0.003
		Omnivore/predators (7), Fungi (7)	0.544	0.008	

* Figures in brackets denote the number of months after incorporation that the parameter was measured.

Suppression of lesion nematode

Analysis of variance showed significant amendment effects on most parameters measured, some effects of inoculating with *Pratylenchus*, but no interactions (Table 6). Pots to which *Pratylenchus* was added had fewer bacterial and fungal-feeding nematodes than non-inoculated pots. Soils amended with sawdust + N, sugarcane trash + N and grass hay + N generally had more fungal-feeding and omnivore-predator nematodes than soils amended with lucerne hay + N or mill mud. Grass hay + N reduced numbers of *Tylenchorhynchus* and *Pratylenchus* in soil relative to the non-amended control, but all amendments except mill mud reduced numbers of *Pratylenchus* in roots.

The effect was greatest with lucerne hay and grass hay, which reduced nematode densities in roots by more than 90% relative to the non-amended soil. None of the amendments affected the growth of sugarcane.

Table 6—Effect of amending soil 6 months previously with various organic materials, and inoculation with lesion nematode (*Pratylenchus zaeae*), on growth of sugarcane in pots, the number of free-living and plant parasitic nematodes/200 mL soil, and numbers of *Pratylenchus*/g root.

Treatment	Log (number of nematodes/200 mL soil +1)										Log no. <i>Pratylenchus</i> /g root	DW tops (g)	DW roots (g)	
	Fungal feeders		Bacterial feeders		Omnivores and predators		Tylenchorhynchus		Pratylenchus					
Nil	2.56	bc	2.71	a	1.52	bc	1.75	bc	2.18	bc	3.04	d	5.7	2.6
Sawdust +N	3.00	d	3.21	c	2.24	d	1.30	ab	1.77	b	2.39	b	6.1	3.2
Cane trash +N	2.72	cd	2.80	a	1.97	cd	1.62	ab	1.98	b	2.63	bc	5.2	2.4
Grass hay +N	2.44	bc	2.62	a	2.09	cd	1.15	a	1.09	a	1.60	a	5.7	2.7
Lucerne hay +N	2.07	a	3.10	bc	0.37	a	1.76	bc	1.84	b	1.95	a	5.3	2.3
Mill mud	2.34	ab	2.87	ab	1.12	b	2.28	c	2.42	c	2.80	cd	6.7	2.7
Lsd (P= 0.05) (Amendment)	0.332		0.284		0.589		0.586		0.431		0.391		n.s.	n.s.
+ <i>Pratylenchus</i>	2.37	x	2.74	x	1.45		1.57		1.94		2.56		6.03	2.72
- <i>Pratylenchus</i>	2.67	y	3.04	y	1.65		1.72		1.82		2.24		5.51	2.57
Lsd (P = 0.05) (<i>Pratylenchus</i>)	0.192		0.163		n.s.		n.s.		n.s.		n.s.		n.s.	n.s.

Means in the same column and treatment group that are followed by the same letter are not significantly different (P = 0.05).

Discussion

The addition of organic materials to soil had major effects on soil chemistry. The high-N amendments (lucerne hay, feedlot manure, poultry manure, chitin and mill mud) had the largest impact, decreasing the C/N ratio of amended soils and also decreasing pH and increasing EC 1, 4 and 7 months after incorporation. In contrast, sawdust, sugarcane trash and grass hay had little effect on these parameters. The C/N ratios of the soil in all treatments declined with time and as this occurred, increasing amounts of NO₃ -N were mineralised. By 7 months, soils amended with nitrogenous materials contained high concentrations of NO₃- N, and net mineralisation had commenced in sawdust, sugarcane-trash and grass-hay treatments.

Microbial biomass and populations of bacteria and fungi increased in most amended soils, but these effects were mainly apparent at 1 and 4 months. By 7 months, microbial populations in amended and non-amended soils were similar, probably because readily utilisable sugars and carbohydrates had been consumed. It is also likely that microbial populations in amended soils were being kept in check at this time by the nematodes and other organisms that feed on microorganisms (Trofymow and Coleman, 1982). Addition of supplemental nitrogen to materials with a high C/N ratio had little long-term impact on the soil biology, but microbial biomass and microbial enzyme activity tended to increase more quickly when additional nitrogen was added. Some amendments appeared to have a temporary detrimental effect on microbial functioning (enzyme activity), notably feedlot manure and poultry manure, but these effects had dissipated by 7 months.

Nematodes are often used as biological indicators (Wasilewska, 1997; Yeates and Bongers, 1999) and our study demonstrated that they provided a good indication of the biological changes induced by various organic amendments. Initially, the nematode community in amended soils was dominated by opportunistic bacterial-feeding species that have relatively short life cycles and high reproductive rates. They are the colonisers in the ecological succession scale of Bongers (1990), and they quickly increase in abundance when organic matter is added to soil. Once readily available nutrients are exhausted, fungi become the most important component of the detritus food web, as they are able to utilise the more recalcitrant fractions of the organic matter such as polysaccharides and lignins. Fungal-feeding nematodes therefore increase in population density as decomposition proceeds, as has been observed previously in studies of decomposing barley straw and other organic materials (Wasilewska *et al.*, 1981; Sohlenius and Bostrom, 1984). The persisters (omnivores and predators with long life cycles and limited colonising ability) take much longer to multiply, but by 7 months they were a significant component of the nematode community, particularly in soils amended with sawdust, sugarcane trash and grass hay.

The chemical composition of the amendments had major effects on the nematode community. Nitrogenous materials favoured bacterial-feeding species while materials with a high C/N ratio favoured fungal-feeders. This suggests that decomposition pathways were dominated by bacteria when supplies of nitrogen were adequate and by fungi when nitrogen was more limited. Similar observations were made by Sohlenius and Bostrom (1986). Interestingly, populations of omnivorous nematodes (*Dorylaimida*) increased in soils amended with high C/N materials and decreased when nitrogenous materials were added. These relatively large nematodes occupy a relatively high trophic level within the food web and are usually not abundant in cultivated soils because they are susceptible to environmental disturbance. They may have been affected by the high concentrations of NO₃-N present in soils amended with large quantities of nitrogenous materials.

Two assays were used to measure suppression to root-knot nematodes and our data suggested that treatments were less suppressive when they were compared using the plant rather than the soil assay. However, this difference may have been due to the arithmetic nature of the gall-rating system used in the plant assay. The level of galling was rated on a scale of 0–10, but for every increase of 1 in the gall rating, there is a logarithmic increase in the number of nematodes in roots. Thus, a difference of one gall-rating unit relative to sand showed in the data as a 10% reduction in galling. However, it represented a much larger reduction in the number of nematodes in the root system.

Results from the plant assay at 7 months suggested that the suppressiveness of soil to root-knot nematode was only enhanced by materials with a high C/N ratio. However, the soil assay showed that all amendments except mill mud made soil more suppressive. The pot experiment produced somewhat similar results, as mill mud did not induce suppressiveness to plant-parasitic nematodes whereas all plant-derived materials reduced numbers of lesion nematode in roots. Sugarcane grown in soil amended six-months previously with lucerne hay + N, grass hay + N, sugarcane trash + N or sawdust + N had 53%–95% fewer lesion nematodes in roots than plants growing in non-amended soil. The reasons for the different responses in the different assays are not known, but it is possible that some amendments enhance biological suppressiveness in soil but also increase the susceptibility of plants to attack by nematodes.

One month after the organic materials were incorporated, the suppression indexes for non-amended soil in the soil and plant assays were 0.44 and 0.74, respectively. This indicates that the

soil was partially suppressive to root-knot nematodes. Non-amended soil then became less suppressive with time and after 7 months it behaved in both assays in much the same way as the pasteurised sand used as a standard. Thus the increased suppressiveness of amended soils after 7 months was due in part to a reduction in the rate of decline in suppressiveness that occurs naturally when soil is bare fallowed.

We made no attempt to identify the nematode-suppressive mechanisms that were enhanced by the addition of organic matter. However, our results clearly show that soils with low levels of nitrate nitrogen, a fungal-dominant biology and high numbers of omnivorous nematodes were most suppressive to plant-parasitic nematodes. Since the omnivorous Dorylaimida are known to prey on nematodes or their eggs (Yeates *et al.*, 1993), they may have been one of the suppressive forces. It is also likely that parasitic and predatory fungi were involved, as they are important biological control agents of nematodes (Stirling, 1991). The predatory hyphomycetes and several genera of wood-decaying basidiomycetes are commonly found in habitats that are rich in cellulose and lignin and are thought to have evolved the capacity to obtain additional nitrogen by capturing nematodes (Barron, 1992; Tzean and Liou, 1993). When materials such as sawdust, sugarcane trash and grass hay are added to soil, these fungi may multiply on free-living nematodes and coincidentally capture plant-parasitic species.

From a practical point of view, our work demonstrates that once a biologically suppressive soil is established, nematode control is maintained for at least 7 months. This is longer than is achievable with organophosphate and carbamate nematicides, as they degrade relatively quickly in soil (Johnson *et al.*, 1981) and inhibit rather than kill nematodes (Bunt, 1987). Our results also provide important insights into how organic inputs might be managed to enhance nematode control in a sugarcane cropping system. We found that amended soils were most conducive to plant-parasitic nematodes when the soil food web was dominated by bacteria. Since the decomposition of legume residues results in such a food web (Stirling *et al.*, 2002), this may explain why populations of pest nematodes increase rapidly when sugarcane is planted following soybean. We also demonstrated that soil is most suppressive to plant-parasitic nematodes when the decomposition pathway is dominated by fungi, and that a food web of this nature can be produced by adding organic matter with a high C/N ratio to soil. The crop residues remaining in the field after the final sugarcane harvest is an obvious source of such material. By manipulating the time and method of incorporating sugarcane residue and the residues from leguminous rotation crops, it may be possible to shift the soil biology in the desired direction. Further studies should therefore focus in this area. Of particular interest is the performance of sugarcane after it is planted in amended soils, as the level of net nitrogen mineralisation in fungal-dominated soil food webs is affected by the C/N ratio of the organic amendment (Chen and Ferris, 1999).

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