COMBINED EFFECT OF NEMATODES AND RATOON STUNTING DISEASE ON SUGARCANE

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Abstract

A field trial was established to investigate the possible interaction between nematodes and the bacterium *Clavibacter xyli* subsp *xyli*, the causal agent of ratoon stunting disease (RSD) in sugarcane. The trial site was a sandy soil infested with species of *Pratylenchus, Meloidogyne Paratrichodorus* and *Xiphinema*. The trial was established with RSD-infected and RSD-free sugarcane variety N12 and the nematodes were controlled with Temik. Samples taken shortly before the plant crop was harvested showed that 10% of the stalks of the initially healthy cane had become infected, whereas 45% of stalks in the diseased plots were infected. Comparable data for the first ratoon crop were 6 and 52%. The higher level of infection resulted in a reduction in sucrose yield of 12% and 23% in the plant and ratoon crop respectively. Much greater losses were caused by the nematodes. In both the plant and first ratoon crops the combined effect of a heavy infection with *C. xyli* subsp *xyli* and large numbers of nematodes on the yield of cane and sucrose was additive.

Introduction

Species of plant-feeding nematodes of the genera *Meloidogyne, Paratrichodorus, Pratylenchus* and *Xiphinema* are important constraints on sugarcane production on the sandy soils in South Africa (Spaull and Cadet, 1990). Estimated losses in 1990 exceeded 600 000 tons cane per annum, equivalent to about 3% of the total crop (Spaull *et al*, 1990).

Ratoon stunting disease (RSD) is a widespread and important disease of sugarcane. In South Africa, losses may exceed 2% of the annual crop (Bailey and Tough, 1991). The causal agent of the disease, *Clavibacter xyli* subsp. *xyli*, is spread between fields mainly by planting infected seedcane, and within fields by harvesting implements, which carry infected juice from diseased to healthy stalks (Bailey and Tough, 1992b). Recent data show that spread of the disease can occur also when healthy seedcane is planted in land that previously carried an infected crop (Bailey and Tough, 1992a). The bacteria survive in the soil and presumably infect newly planted sugarcane via the roots. Nematodes feed on the roots and may therefore facilitate the entry of the bacteria. In addition, both *C. xyli* subsp *xyli* and species of *Paratrichodorus* and *Xiphinema* affect stalk elongation in sugarcane by impairing water movement into or through the plant. The nematodes restrict uptake of water by damaging the roots (Cadet and Spaull, 1985; Spaull and Cadet, 1991) and the bacteria restrict movement of water through the plant by blocking the xylem vessels (Gillaspie and Teakle, 1989). Thus it might be expected that where the bacterium and plant-feeding nematodes occur together, there may be an interaction. However, investigations in Brazil showed that *C. xyli* subsp *xyli* did not interact with such nematodes in sugarcane, the effect of the two together being additive, although these studies were on plant cane only (Anon, 1977; Regis and Moura, 1989).

In the present study the combined effect of nematodes and RSD on the yield of sugarcane was measured in the plant crop and in the first ratoon.

Methods

The trial was conducted at the Experiment Station farm at La Mercy on the plant and first ratoon crop of sugarcane variety N12. This variety is becoming increasingly popular in the rainfed areas of production and constituted 25% of the crop crushed in these areas in 1992. The soil was a deep structureless fine sand of the Fernwood form with 4% clay and 4% silt.

Five weeks before the trial was established, the regrowth of the previous crop was sprayed with Roundup (35% glyphosate) at 8 l/ha. A week later the stools were ploughed out and then disced twice over a period of two weeks. It was later found that parts of a number of stools survived this treatment. These were removed by hand.

The trial plots, each comprising five rows 12 m long (3 rows × 8 m net plot) with a 1,2 m spacing, were planted with healthy and RSD-infected seedcane. The two groups of seedcane were identical except that the infected cane was from a propagation plot that had been planted with setts inoculated with juice expressed from RSD-infected stalks.

The trial was planted in March 1990 and the plant crop was harvested after 15,3 months, in June 1991. The following ratoon was harvested after 13,6 months, in August 1992.

To check the level of infection of the cane by *C. xyli* subsp *xyli*, a 20 stalk sample was collected per plot, shortly before each of the two crops was harvested. The xylem sap was expressed from the stalks and examined microscopically (Bailey and Fox, 1984). This showed that 10% of the stalks of the initially healthy plant cane had become infected with *C. xyli* subsp *xyli*, whereas 45% of the stalks in the diseased plots were infected. Comparable data for the first ratoon crop were 6% and 52%. To limit the spread of infection, the initially healthy plots were harvested with knives cleaned with a 10% solution of Jeyes Fluid (carbolic acid) (Bailey and Tough, 1992b). After cutting every four to six stalks, the knives were dipped in this solution. No attempt was made to clean the knives when the plots of heavily infected cane were harvested.

Details of the treatments imposed on the lightly, and on the heavily infected cane are given in Table 1. The trial was arranged in randomised blocks with six replicates per treatment. Treatment No. 4 in the ratoon crop was to assess the carry-over effect of the high rate of Temik applied to the plant crop.

To monitor the effect on the nematodes of treatments with the high rate of Temik, soil samples were taken on three occasions from treatments 1 and 4 in the plant crop (see Table 1). They were taken also on four occasions from treatments 1 and 2 in the ratoon crop. Each sample comprised 20 cores per plot to a depth of 220 mm, and the nematodes

Table 1

Treatments applied to the plant crop, and to the same plots in the following ratoon crop, of cane lightly infected with RSD and cane heavily infected with RSD

Treatment number	Plant crop	Ratoon crop
1	Untreated control	Untreated control
2	Untreated control	Temik 150G at 77 kg/ha applied over the row as a split application, half at 9 weeks and half at 18 weeks after harvest.
3	Temik 150G at 20 kg/ha applied in the furrow at planting	Temik 150G at 20 kg/ha applied over the row 9 weeks after harvest.
4	Temik 150G at 60 kg/ha applied as a split application, half in the furrow at planting and half 4 weeks later	Untreated.

were extracted by the decanting-sieving method of Brown and Boag (1988). At harvest the number, length and mass of stalks in the three, 8 m long net rows were recorded, and a 12 stalk sample was processed from each plot, to determine the mass and proportion of sucrose in the cane.

Results

Four species of endoparasitic nematodes were recorded in the trial site, viz. *Meloidogyne incognita, M. javanica, Pratylenchus zeae* and, infrequently, *Rotylenchulus parvus.* There were eight species of ectoparasites, viz. *Helicotylenchus dihystera, Paratrichodorus lobatus, Paratrichodorus* sp, *Xiphinema elongatum* and, infrequently, *X. vanderlindei, Paralongidorus* sp, *Neodolichodorus* sp and *Criconemella* sp.

In both the plant and ratoon crops, good control of the nematodes was achieved within four or five weeks after treatment with the high rate of Temik (Table 2). By the time the plant crop was harvested the populations had recovered, but in the following ratoon the effect of treatment on the nematodes persisted through to harvest (Table 2).

Table 2

Number of nematodes per 100 ml soil in plots treated with the high rate of Temik (T) and in untreated plots (C) at various times after planting and after harvesting the plant crop. Figures represent means from plots of cane lightly and heavily infected with RSD

		Plant crop			First ratoon crop			
		Weeks after planting			Weeks after harvest			
		0	4	49	3	14	20	55
Pratylenchus	C	29	15	19	62	33	21	10
	T		6	23	56	11	1	3
Meloidogyne	C	5	3	85	22	15	41	3
	T		0	54	23	13	1	1
Helicotylenchus	C	80	35	91	74	69	101	49
	T		11	148	79	19	3	9
Xiphinema	C	71	39	68	94	65	80	70
	T		17	86	83	23	7	28
Paratrichodorus	C	31	14	78	36	55	42	7
	T		6	75	38	9	6	3

In the untreated plots of the plant crop, numbers of nematodes present after 49 weeks were about the same or more than those present when the crop was established (Table 2). In the first ratoon there were fewer nematodes at the end of the crop.

Damage to the roots was observed two months after planting. There were galls on the sett roots caused by *Meloidogyne* species. In addition there was severe damage to the shoot roots, probably caused by the species of *Xiphinema* and *Paratrichodorus*, viz. the root system was sparse and the lateral roots were short and stunted (see Figure 3 in Spaull and Cadet, 1990). Similar damage to the shoot roots was present in the untreated plots of the following ratoon crop.

Table 3

Effect of Temik on some yield components of the plant crop of cane lightly infected with RSD (L. inf) and on cane heavily infected with RSD (H. inf). Figures in parenthesis represent the differences in yield expressed as a percentage of that of the lightly infected cane. Figures with a double asterisk are signficantly greater than the untreated control (p < 0,01)

Treatment number	Treatment (plant crop)	Level of RSD	Sucrose t/ha	Ers % cane	Stalks (1 000/ha)	Stalk length (cm)
1 and 2	Control	L. inf	5,1	11,0	88	127
		H. inf	4,5 (−11%)	11,1	95 (+8%)	118 (−7%)
3	Temik, 20 kg/ha	L. inf	10,1**	12,7**	106**	160**
		H. inf	9,3** (−8%)	13,0**	106** (0)	154** (−4%)
4	Temik, 60 kg/ha	L. inf	15,2**	13,2**	124**	194**
		H. inf	12,9** (−15%)	13,2**	122** (−2%)	177** (−9%)
LSD p < 0,05			2,4	0,7	11,2	24,1
LSD p < 0,01			3,2	1,0	15,0	32,3
Mean, all treatments		L. inf	8,9	12,0	102	152
		H. inf	7,8 (−12%)	12,1	104	142 (−7%)
LSD p < 0,05			1,2	0,4	5,6	12,0
Mean Control			4,8	11,0	91	123
Mean Temik 20 kg/ha			9,7**	12,9**	106**	157**
Mean Temik 60 kg/ha			14,0**	13,2**	123**	185**
LSD p < 0,05			1,7	0,5	7,9	17,0
LSD p < 0,01			2,3	0,7	10,6	22,8

Table 4

Effect of Temik on some yield components of the first ratoon crop of cane lightly infected with RSD (L. inf) and of cane heavily infected with RSD (H. inf). Figures in parenthesis represent the difference in yield expressed as a percentage of that of the lightly infected cane. Figures with a double asterisk are signficantly greater than the untreated control or the appropriate mean value, p < 0,05 and p < 0,01 respectively

Treatment number	Treatment (ratoon crop)	Level of RSD	Sucrose t/ha	Ers % cane	Stalks (1 000/ha)	Stalk length (cm)
1	Control	L. inf	3,8	13,7	84	80
		H. inf	2,2 (−42%)	13,5	69 (−17%)	68 (−15%)
3	Temik, 20 kg/ha	L. inf	8,6**	14,6	102*	123**
		H. inf	7,1** (−17%)	14,6*	96** (−6%)	118** (−4%)
2	Temik, 77 kg/ha	L. inf	12,3**	14,7*	123**	149**
		H. inf	10,2** (−17%)	14,3	110** (−10%)	138** (−8%)
4	Residual Temik	L. inf	4,8	14,3	87	92
		H. inf	3,4 (−29%)	14,0	77 (−11%)	75 (−18%)
LSD p < 0,05			1,8	0,9	13,6	13,8
LSD p < 0,01			2,5	1,2	18,2	18,5
Mean, all treatments		L. inf	7,4**	14,3	99**	111**
		H. inf	5,7 (−23%)	14,1	88	100 (−10%)
LSD p < 0,05			0,9	0,4	6,8	6,9
LSD p < 0,01			1,2	0,6	9,1	9,2
Mean Untreated Control			3,0	13,6	77	74
Mean Temik 20 kg/ha			7,9**	14,6**	99**	121**
Mean Temik 77 kg/ha			11,3**	14,5**	117**	144**
Mean Residual Temik			4,1	14,2	82	84*
LSD p < 0,05			1,3	0,6	9,6	9,7
LSD p < 0,01			1,7	0,8	12,9	13,1

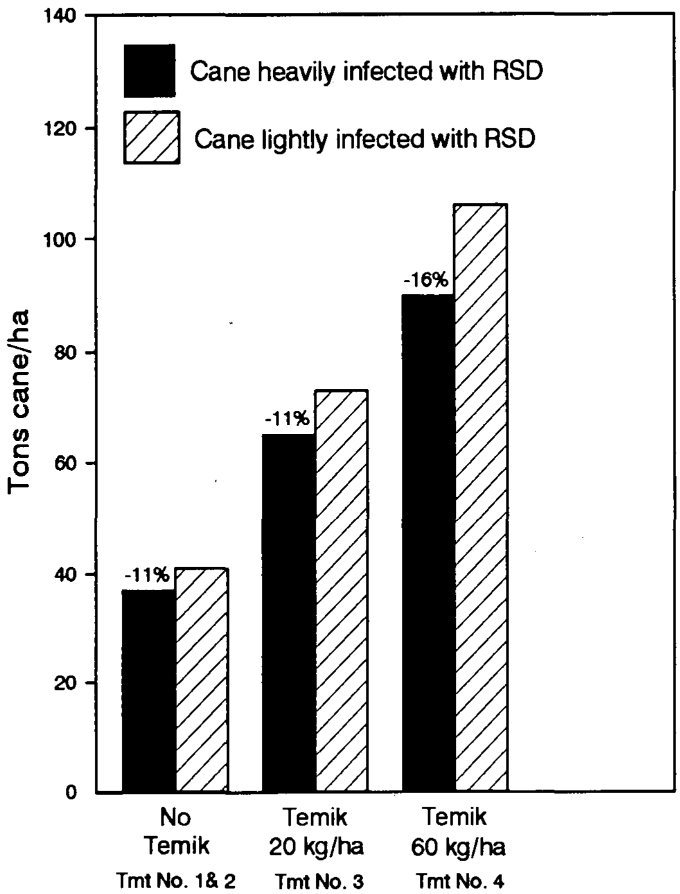

FIGURE 1 Effect of Temik on the yield of the plant crop of cane lightly infected with RSD and cane heavily infected with RSD (t cane/ha).

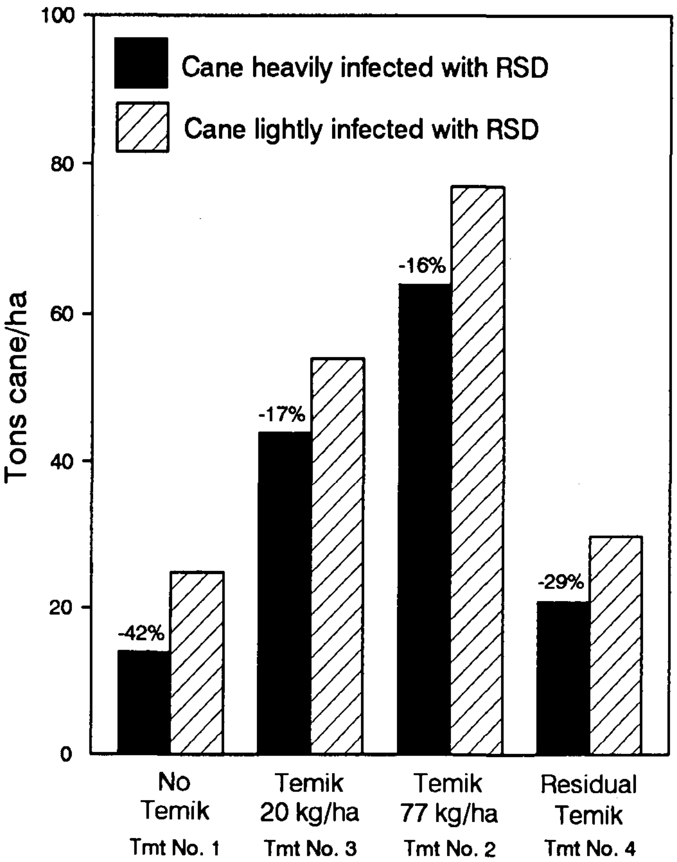

FIGURE 2 Effect of Temik on the yield of the first ratoon crop of cane lightly infected with RSD and of cane heavily infected with RSD (t cane/ha).

Judging from the response to treatment with Temik, nematodes had a marked effect on the yield of cane in both the plant crop and the first ratoon ($p < 0,01$) (Tables 3 and 4; Figures 1 and 2). The response of the cane to treatment was greater in the first ratoon than in the plant crop, and the response to the higher rates of Temik was much greater than the response to the lower rate (Tables 3 and 4; Figures 1 and 2). Thus, in the plant crop, the yield of cane treated with 20 kg Temik/ha was on average 76% greater than the untreated cane. In the following ratoon the yield was 161% greater than the control. Comparable figures for the response to treatment at 60 kg Temik/ha in the plant crop, and 77 kg/ha in the ratoon, were 151% and 275% respectively (Figures 1 and 2). There were similar responses to treatment with Temik when yield was measured in tons of sucrose (Tables 3 and 4).

Treatment with 60 kg Temik/ha in the plant crop had no significant effect on yield in the following ratoon (Treatment No. 4 in Table 4 and Figure 2).

The responses that followed treatment with Temik were a result of an increase in the number, length and mass of stalks as well as an increase in the amount of recoverable sucrose in the cane ($p < 0,01$) (Tables 3 and 4).

In both the plant and ratoon crops, the highest yield occurred in nematicide treated plots with the lightly RSD-infected cane (Tables 3 and 4; Figures 1 and 2). The lowest yields occurred in plots of untreated, heavily infected cane. The reduction in yield of this cane was about the same as the combined reduction in yield of the nematicide treated, heavily infected cane plus that of the untreated, lightly infected cane (Table 5).

Table 5

Reduction in yield of sugarcane from the various treatments as a percentage of the yield of cane lightly infected with RSD and treated with Temik

Treatment	Amount of Temik applied to treated plots (kg/ha)			
	Plant crop		Ratoon crop	
	20	60	20	77
Temik, cane lightly infected	100	100	100	100
Temik, cane heavily infected	-11	-16	-17	-16
Control, cane lightly infected	-43	-61	-53	-67
Control, cane heavily infected	-49	-65	-73	-81

In the plant crop, the yield of the untreated or the nematicide treated, heavily infected cane was 11 or 16% less than that of the lightly infected cane (Figure 1). However, in the following ratoon crop, while the yield of the heavily infected cane was reduced by about 17% where a nematicide was used, the yield of the corresponding cane that had never been treated was reduced by 42% (Figure 2). In the plots that were treated with Temik in the plant crop but not in the ratoon (treatment No. 4), the yield of heavily infected cane was 29% less than that of the lightly infected cane (Figure 2).

As before, comparable responses occurred when yield was measured in tons of sucrose (Tables 3 and 4).

In the plant crop, there was no significant difference between the yield components of the cane lightly infected with RSD and the heavily infected cane (Table 3). However, in the ratoon crop, mass of cane and sucrose and number and length of stalks were all greater in the lightly infected cane ($p < 0,01$) (Table 4). In neither crop did the level of infection with RSD affect cane quality as measured by the proportion of recoverable sucrose in the stalks.

The plant crop received 1 498 mm rainfall, which represents 120% of the long term mean (LTM). Eighteen per cent of the rain fell during the first month after planting. In contrast, the ratoon crop received only 653 mm rain (61% LTM). Water use, as measured by the yield in tons cane per hectare per 100 mm rainfall, was more efficient in the ratoon cane treated with a nematicide than in the other treatments (Table 6). Water use efficiency was least in the untreated ratoon cane that was heavily infected with RSD.

Table 6

Yield of sugarcane per 100 mm rain for the plant and ratoon crops of cane lightly infected with RSD (L. inf) and cane heavily infected with RSD (H. inf)

Treatment		Plant crop Total rain = 1 498 mm	Ratoon crop Total rain = 653 mm
		t cane/ha/100 mm	t cane/ha/100 mm
Control	L. inf	2,7	3,8
	H. inf	2,5	2,2
Temik 20 kg/ha	L. inf	4,8	8,3
	H. inf	4,3	6,8
Temik 60 or 77 kg/ha	L. inf	7,1	11,8
	H. inf	6,0	9,9

Discussion

Infection of the initially healthy cane was presumably caused by *C. xyli* subsp *xyli* that survived in the soil or in old roots after the attempt had been made to eradicate the previous crop. Subsequent to the planting of this trial, and based on other work, Bailey and Tough (1992a) recommended that, instead of replanting fields within a few weeks of destroying the old crop, as is commonly done in South Africa, a break of six months is advisable if the old crop is infected with RSD.

Since the unintended infection in the intended healthy plots was much lighter than that in the deliberately infected cane (cf. 6-10% with 45-52%), meaningful comparisons can still be made between lightly infected and heavily infected plots. The reductions in yield of recoverable sucrose, caused by RSD in the plant and first ratoon crops of 12% and 23% (mean of all treatments, Tables 3 and 4) are similar to those reported previously for N12 under rainfed conditions (Bailey and Bechet, 1986).

Treatment with the high rates of Temik gave a marked reduction in the number of nematodes, compared with the untreated controls, but did not eliminate them. There were thus two densities of nematodes, a relatively high level of infestation and a low one. In addition there was probably a third, intermediate, level as treatment with Temik at 20 kg/ha is less effective in reducing the number of nematodes associated with sugarcane than treatment at higher rates (unpublished data).

Data from both the plant and ratoon crops indicate that when the nematodes and *C. xyli* subsp. *xyli* occur together at high levels, their effect on yield is additive (Table 5). The combined effect of the supposed intermediate density of nematodes, from the treatment with 20 kg Temik/ha, and a high level of *C. xyli* subsp. *xyli* was also additive (Table 5).

These observations are in agreement with previous reports on the association between RSD and nematodes on plant cane in Brazil (Anon, 1977; Regis and Moura, 1989). However, it is worth noting the disproportionate reduction in cane yields associated with the high level of RSD in the untreated ratoon cane that was also untreated in the plant crop (-42%) and, to a lesser extent, in the untreated ratoon cane that had previously been treated with the high rate of Temik (-29%) (Figures 1 and 2). In the other treatments the high level of RSD reduced yields by 11-17%. Further data are required to clarify the relationship.

In the plant and ratoon crops, treatment with Temik at 60 and 77 kg/ha increased yields above those produced from treatment with 20 kg/ha; which in turn, increased yields above those of the untreated control. As stated earlier, the response to the higher rate was presumed to be a consequence of better nematode control. However, it could be argued that since aldicarb, the active ingredient of Temik, can enhance the growth of some plants in the absence of pests (Garabedian and van Gundy, 1982; Barker *et al*, 1988; Barker and Powell, 1988) the response of the cane to treatment at all three rates was due more to growth stimulation by the chemical than to nematode control. However, the results of a pot trial conducted in 1979 suggest that this is not the case. In the trial Temik 150G was applied to recently planted setts of NCo376 in sterile sand at rates of 11,7, 23,3 and 46,7 kg/ha. After 11 weeks of growth, the highest rate depressed the mass of the roots by 25%, and at the two higher rates, scorching of the leaf tips was significantly greater than in the untreated control ($p < 0,05$). Shoot mass and nutrient content of the leaves were not affected by any of the treatments (Anon, 1980). The rates of application used in the field trial were greater than, or within the range tested in the pot trial. It therefore seems unlikely that the growth of cane was stimulated by the chemical; rather, the higher rates may have depressed growth. However, several factors may affect the growth response of plants to aldicarb, including the dosage, soil moisture content, soil texture and crop cultivar (Barker *et al*, 1988), so growth stimulation cannot be entirely ruled out.

The results support previous observations that, on poor sandy soils, nematicides are required to sustain yields of sugarcane (Anon, 1982). The marked decline in yield of the untreated ratoon crop, whether treated or not in the plant crop, was a consequence of root damage caused by nematodes, compounded by RSD and, presumably, the very low rainfall (Table 6). In most instances, where moisture is limiting, RSD has a greater effect on cane than when no such stress is present, and the effect of RSD on ratoons is usually greater than that on plant crops (Gillaspie and Teakle, 1989). This would seem to be the case in the present trial, but growing conditions for the two crops were very different.

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