SERRATIA ENTOMOPHILA FOR CONTROL OF GRASS GRUB IN STRAWBERRIES

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SUMMARY

The bacterium, *Serratia entomophila,* was applied to a strawberry crop infested with grass grub. Applications were made either through the trickle irrigation system or by subsurface injection to individual plants. Soil samples taken 2 and 30 days after application through the trickle system showed bacteria had established in soil. Thirty days after application at 200 litres/ha an average of 12% of larvae were diseased, with lower levels of disease found at 100 and 20 litres/ha. Treatment of individual plants with two rates of bacteria resulted in average disease levels of 42 and 11%. Applications to pots gave a disease level of 67%.

INTRODUCTION

Grass grub, *Costelytra zealandica,* can be a significant pest of strawberries and other small fruits (Scott 1984). Larvae feed on the roots, removing fine feeding roots and, in some cases, woody roots at the base of the crown. Plants appear stunted and have a shorter cropping life than uninfested plants. Recommended treatments are usually preventative: persistent insecticides such as lindane and DDT are applied to soil before mounding (Langford 1987). When larvae are present under polythene control is difficult. If the infestation is limited, application of insecticides to individual plants may be warranted (Scott 1984).

Concern over the use of persistent chemicals has led to investigation of alternative methods for pest control. *Serratia entomophila* has been used to control grass grub in pasture (Jackson *et al* 1986). In this study, pot and field trials were conducted to investigate the potential use of *S. entomophila* for control of grass grub in strawberries.

MATERIALS AND METHODS

Culture method

S. entomophila strain A1 was originally isolated from a diseased grass grub larva collected from pasture at Fairton, Canterbury. Bacteria required for application were produced by liquid culture in a 1000 litre fermenter at NZAEI, Lincoln. Cultures were stored at 4 °C and contained at least 2×10^9 bacteria/ml at time of application.

Pot trial

A pot trial was conducted using young strawberry plants and soil collected from the trial site. Ten healthy larvae were added to each of 10 replicate pots when plants had become established. Five pots were then inoculated with bacteria by surface application at a rate of 200 litres/ha. The pots were destructively sampled 30 days after inoculation and larvae counted and examined for disease symptoms.

Field application

Field trials were conducted in February 1988, on a 4 year old block of strawberries on a horticultural property near Christchurch. Strawberries were growing on raised beds under polythene. The soil was a Waimakariri sandy loam and was found to contain no *S. entomophila.* A preliminary population sample was conducted to determine the distribution of grass grub throughout the field. Twenty plants and soil surrounding their roots were collected at random from the trial area and larvae counted.

Bacteria were introduced into 100 m rows by attaching a Cambrian hand pump pressure sprayer (1 litre) to the trickle irrigation pipe. As plants varied in vigour, treatment rows were graded on a scale of 1 - 4 (1 = healthy plants, 4 = plants less vigorous and starting to senesce). Treatments were allocated to rows so variations in

plant vigour were distributed across treatments. Three rates of bacteria were applied: 200, 100 and 20 litres/ha, with four replicate rows of each rate. Weather conditions at the time of application were overcast and the soil moist (25% soil moisture).

The distribution of bacteria immediately after application was examined by collecting 10 soil cores (25 × 100 mm) from soil adjacent to the trickle lines and 100 mm away from the lines in rows treated with 200 litres/ha. Soil was also sampled for the presence of *S. entomophila* at 2 and 30 days after application when 20 soil cores were collected from the centre of each row. Samples were pooled and plated on an agar selective for *Serratia* spp. and incubated for 4 days at 30 °C. Colonies were counted and identified by further biochemical tests. Fourteen and 30 days after application, four plants and surrounding soil were removed from each treatment row. Larvae were counted and assessed for disease. No further sampling to determine mortality was possible as the block was destroyed by cultivation.

In the same block, subsurface applications were made by injecting bacterial solution into four holes around the crown of individual plants. Two rates were compared: 4 × 2 ml and 4 × 5 ml (1.6×10^{10} and 4×10^{10} bacteria/plant). Fifty plants were treated with each rate. Thirty days after application, 10 plants were sampled from each treated and untreated row and larvae were assessed for disease symptoms.

RESULTS

Pot trial

Application of bacteria to pots resulted in a mean disease level of 67% (SE = 13). No disease was found in untreated pots in which healthy larvae had removed all fine feeding roots. There was less root damage in pots where bacteria had been applied. Control pots containing no larvae allowed good root growth by the plants.

Field application

In the population sample taken prior to treatment, the number of larvae beneath individual plants ranged from 0 - 30, with a mean of 9 per plant. Larvae were healthy second and third instars and were found mainly within the root mass. In most cases all fine feeding roots had been removed and only woody roots remained.

Immediately after application high numbers of *S. entomophila* were recovered from soil adjacent to the irrigation lines (mean $\log_{10}$ number of 6.58 /g oven dry soil). *S. entomophila* was not recovered from soil 100 mm from the lines. Mean log numbers of bacteria in the soil 2 and 30 days after application at each rate are shown in Table 1. The numbers of bacteria recovered increased with higher rates of application.

TABLE 1: Bacterial numbers ($\log_{10}$/g oven dry soil) remaining in soil 2 and 30 days after application at three rates.

Rate of bacteria (litres/ha)	Numbers of bacteria in soil	
	2 days	30 days
20	4.06	3.41
100	4.85	3.90
200	4.88	4.36
Untreated	(0.00)	(0.00)
SED	0.380	0.397
Significance of trend	*	*

Disease levels resulting from application along trickle lines are shown in Table 2. Square root transformation of data was used to assist homogeneity, before analysis of variance was performed. Thirty days after application, there was a significant response to rate of treatment ($P < 0.01$). At 200 litres/ha, an average of 12% of larvae were diseased. Lower numbers of diseased larvae were found after treatment with 100 and 20 litres/ha. No disease was found in untreated plots.

Treatment of individual plants resulted in higher percentages of infected larvae (Table 3). At the highest rate of application an average of 42% of larvae were diseased. Treatment with the low rate of bacteria averaged 11% disease incidence.

TABLE 2: Percentage of diseased larvae (sq rt transformed) recovered 14 and 30 days after treatment with three rates of bacteria. (Back transformed means are shown in brackets).

Rate of bacteria (litres/ha)	Percentage larvae infected 14 days	30 days
20	1.3 (1.6)	1.1 (1.2)
100	3.0 (8.9)	2.3 (5.4)
200	2.7 (7.2)	3.5 (12.2)
Untreated	(0.0)	(0.0)
SED	1.05	0.65
Significance of trend	n.s.	**

TABLE 3: Percentage of diseased larvae (sq rt transformed) recovered 30 days after treatment of individual plants with two rates of bacteria. (Back transformed means are shown in brackets)

Rate of bacteria (ml/plant)	Percentage larvae infected
4 × 2 ml	3.31 (10.9)
4 × 5 ml	6.52 (42.5)
Untreated	(0.00)
SED	2.52
Significance	*

DISCUSSION

The results of the pot trial showed that high levels of disease could be induced in the target larvae. In the field, the bacteria were successfully established and survival was high for at least a month. The environment under polythene is ideal for disease development as the soil remains relatively warm and moist. Larvae are clustered in roots under plants so spread of the disease between individuals is possible.

Application through trickle lines resulted in establishment of disease in the grass grub population. Levels of disease, however, were low. This result probably reflects an uneven distribution of bacteria in soil, with high numbers of bacteria found adjacent to the trickle lines, while the majority of the larvae were beneath the crown of the plant. Some movement of the bacteria with irrigation water is likely, but this needs to be determined. Similarly the distribution of bacteria along the rows from point of application should be examined. An even application of bacteria by this method will depend on sound irrigation lines running along the centre of rows.

Applications to individual plants resulted in higher levels of disease, probably because bacteria were placed among the plant roots where most larvae were found. This method may be feasible for treating small localised areas of grass grub, but would not be economic on a commercial property unless a mechanised method was available.

The results of these preliminary experiments show that *S. entomophila* has potential for controlling grass grub in strawberries. Methods and strategies for application require further investigation but with adequate distribution throughout the soil, *S. entomophila* could be used as a curative treatment for grass grub in strawberries.

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