Project title:	**Brassica crops: Evaluation of non-organophosphorus insecticides for controlling the cabbage root fly**
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CONTENTS

PRACTICAL SECTION FOR GROWERS

Commercial benefits of the project

- Without adequate insecticidal control, it is estimated that about 24% of the plants in field brassica crops would be rendered unmarketable by the cabbage root fly.
- In crops such as swedes and turnips (marketed value about £15M/annum), in which the pest attacks directly the part of the crop used for human consumption, the losses would be considerably higher. This sector of the industry may not be sustainable if the cabbage root fly cannot be controlled effectively.
- Even if cultural methods could be relied on to lower overall damage to 15-20%, the Industry could still be facing losses of between £25-33M per annum from the area of crop that needs protecting currently against attacks by the cabbage root fly.

Background and objectives

The cabbage root fly (*Delia radicum*) is the most serious pest of brassica crops in the United Kingdom. Since 1963, the larvae of this pest have been controlled by seed-treatments, drenches, sprays and granular formulations of mainly organophosphorus (OP) insecticides. However, because of public concerns about OPs in particular, DEFRA/PSD are currently reviewing all pesticides with anticholinesterase activity. As a result, some products have been withdrawn already and others may be withdrawn in the future. The need to find alternative methods or insecticides for cabbage root fly control has never been so urgent.

<u>The purpose of this project is to find ways of controlling the cabbage root fly with non-OP insecticides and to find alternative methods of using those compounds which are still available</u>. Three experiments were done in 2001-2: 1) using film-coated seed to kill the eggs and larvae, 2) using insecticide-treated baits to kill the adult flies and 3) establishing whether non-insecticidal treatments could be used to deter female flies from egg laying. Owing to the concern being expressed by swede growers, the experiments concentrated on swede and turnip crops (see Finch, Collier & Jukes, 1999). However, the results of the project apply equally to leafy brassica crops, as levels of control do not have to be as stringent when the pest damages the part of the plant that is not used for human consumption. With leafy brassica crops, once the plants are established, the crop can tolerate some damage to the roots without any measurable loss in yield. In contrast, in swede and turnip crops where the fly larvae damage the part of the plant that is used for human consumption, the crop has to be kept pest-free throughout most of its growth period if the roots are to be acceptable at harvest.

Summary of results and conclusions

- Six insecticides were included in this study, three of which are either OPs or carbamates. In addition, five potential insect deterrents were assessed.
- As a seed treatment, the new microencapsulated formulation of chlorpyrifos (Empire) was no more effective than the standard formulations. Spinosad and chlorfenvinphos both provided some control as seed treatments, but chlorfenvinphos caused phytotoxicity problems. In contrast to the results from previous studies, fipronil and chlorpyrifos appeared to be ineffective.

- When applied in bait solutions, lambda-cyhalothrin, chlorpyrifos, spinosad and fipronil all killed adult cabbage root fly. Fipronil was consistently the most effective insecticide and lambda-cyhalothrin the least effective. Pirimicarb was shown to kill onion fly, but not cabbage root fly.
- None of the non-insecticidal deterrents had any effect on the numbers of cabbage root fly pupae recovered in a field experiment using swedes. Ecoguard (garlic granules) and Seagrow (composted seaweed) showed some deterrent effects in the laboratory, but these effects were not transferred to the field.

Action points for growers

Film-coated seed (field experiment - turnip)

- **Chlorfenvinphos** Quite phytotoxic at higher doses, but it gave some control of cabbage root fly larvae under field conditions.
- **Chlorpyrifos** Phytotoxicity problems can be reduced/eliminated by the addition of an inert filler (Talcum powder). Usually reduces the numbers of cabbage root fly larvae, but was not effective in this trial. Probably reduced numbers of beneficial insects, which appeared to be numerous on control plots.
- **Fipronil** As with chlorpyrifos, it has shown great promise in the past, but in this trial it appeared to be totally ineffective
- **Spinosad** This naturally derived chemical was the most effective in this trial.

Insecticide treated baits (glasshouse experiment)

- **Pirimicarb** Did not kill adult cabbage root flies at the doses tested (but did kill onion flies).
- **Fipronil** The most effective at killing adult cabbage root flies. Nearly 100% mortality of both female and male flies within 24 hours.
- **Spinosad** An effective killer of adult cabbage root flies. Efficacy diminished as residue aged.
- **Chlorpyrifos** Similar in performance to spinosad. Aged residues were almost as effective as fresh ones.
- **Lambda-cyhalothrin** Similar in performance to spinosad and chlorpyrifos against male flies, but less effective against female flies.

Insect deterrents (laboratory and field experiments - swede)

- **Antistress** An acrylic product sold to reduce drought stress and frost damage. Did not deter the cabbage root fly from laying eggs.
- **Ecoguard** A granular garlic extract sold as a micronutrient. Had some deterrent effect against the cabbage root fly, but did not reduce damage in a field trial.

- **Ecospray** A liquid extract of garlic. No evidence of any deterrent effects against the cabbage root fly.
- **Majestik** An oil based starch product which acts as a physical insecticide and is sold to control aphids, spider mites, thrips and whitefly. No evidence of any deterrent effects against the cabbage root fly.
- **Seagrow** A seaweed based organic fertiliser and soil improver. Had some deterrent effect against the cabbage root fly, but increased damage in a field trial.

Anticipated practical and financial benefits

Brassica crops are grown currently on approximately 35,000 ha in the UK and the marketed value of these crops is about £165M/annum [*Basic Horticultural Statistics for the United Kingdom. Calendar and Crop Years 1990/01 – 2000/01. Department for Environment, Food and Rural Affairs, National Statistics*]. In 2002 there are only two approved chemicals, chlorfenvinphos (Birlane) (approval until 25/07/2003) and chlorpyrifos (Dursban), for cabbage root fly control.

Partway through 2003, no product will be available to control the cabbage root fly in swedes and turnips, since chlorpyrifos is not approved on these crops. Hence, the need to find alternatives, particularly for swede and turnip production, has never been greater. As a consequence, the current work has been targeted to look at alternative insecticides, alternative uses for currently approved insecticides, and non-insecticidal alternatives.

SCIENCE SECTION

Introduction to experimental work

The work during this one-year project was "short-term", and was concerned solely with finding possible replacements for the OP-based treatments applied currently. The project involves field and glasshouse trials. Glasshouse trials can be used to estimate how much insecticide is required to give the desired level of control. The advantages of glasshouse trials are that they allow 1) all insecticides to be tested at the same insect pressure and 2) a range of insecticide doses can be tested in a limited space prior to the extensive, and hence more expensive, field trials. In addition, 3) variations in the results caused by changing weather conditions and/or beneficial insects can also be avoided. Results from glasshouse experiments pinpoint directly the treatments unlikely to be accepted in commercial crops. Hence, a strong scientific base can be used to decide which new insecticides to include in new research programmes.

Experiments were done to answer the following 3 questions:

Film-coated seed

1) How effective at controlling the cabbage root fly under field conditions is seed coated with the optimum loading of four test insecticides?

Insecticide treated baits

2) Will any of the currently available insecticides kill adult cabbage root flies if incorporated into one of the commercially available insect baits?

Cabbage root fly deterrents

3) How effective under laboratory and field conditions are five products (Antistress, Ecoguard, Ecospray, Majestik, Majestik and Seagrow) which, it has been suggested, are capable of deterring the cabbage root fly from laying on brassica plants?

The three experiments

For scientific reasons the test chemicals are shown as the active ingredients (with one product name in parenthesis) in the Materials and Methods sections, as certain chemicals are available under a range of different product names.

The actual active ingredients tested, together with the representative product (shown in parenthesis), were: chlorfenvinphos (Birlane), chlorpyrifos (Dursban), pirimicarb (Aphox) fipronil (Fipronil), lambda-cyhalothrin (Hallmark) and spinosad (Spintor)

A. Film-coated seed

Experiment 1. How effective at controlling the cabbage root fly under field conditions is seed coated with the optimum loading of four test insecticides?

a) Choosing the dose for the field trial

Materials and methods

Turnip seeds were film-coated at HRI, Wellesbourne with chlorpyrifos (Gigant), microencapsulated chlorpyrifos (Empire), chlorfenvinphos (75% a.i. microencapsulated formulation), fipronil (experimental seed treatment formulation) and spinosad (Spintor). Both the chlorpyrifos and the chlorfenvinphos formulations were applied by using a slurry with Talcum powder, as both insecticides are liquids at room temperature. Each of the test insecticides was applied at three target loadings, which were based on previous experience with phytotoxicity and seed coat capacities. Chlorpyrifos and chlorfenvinphos were applied at target loadings of up to 50 g a.i./unit (1 unit =100,000 seeds) as phytotoxicity effects were expected above this dose. Fipronil and spinosad were applied at target loadings of up to 80 and 160 g a.i./unit respectively as it was felt that these were the maximum practical loading capacities of the turnip seeds. A PVA sticker at the rate of 0.5 % of product weight was applied with all treatments. The actual loadings achieved were assessed by high performance liquid chromatography (hplc) analysis (Table 1). A further batch of seed was left untreated.

Table 1. Insecticide doses applied to turnip seeds.

Insecticide	Actual dose (g a.i./unit)	(mg a.i./seed)
Chlorfenvinphos	11	0.11
	22	0.22
	50	0.50
Chlorpyrifos (Empire)	8	0.08
	14	0.14
	32	0.32
Chlorpyrifos (Gigant)	8	0.08
	18	0.18
	40	0.40
Spinosad	38	0.38
	79	0.79
	179	1.79
Fipronil	24	0.24
	48	0.48
	80	0.80

On 31 May, the treated turnip seed was sown (14 rows x 22 seeds/treatment) in 308 Hassy trays containing Levington compost. The trays were maintained under glasshouse conditions (20 ± 2 ºC) and the numbers of seedlings that emerged were counted on 11 June (11 days after sowing) and 19 June (19 days after sowing). The results were subjected to Analysis of Variance. The percentage data were angle-transformed prior to analysis.

Results

The results are summarised in Table 2. The highest dose (50 g a.i./unit) of chlorfenvinphos was the only treatment that had a detrimental effect on the numbers of seedlings that ultimately emerged (p=0.05). Chlorfenvinphos (22 g a.i./unit and 50 g a.i./unit) and chlorpyrifos (8 g a.i./unit) all slowed the rate of emergence (p=0.05). Based on this information, the highest dose from each of the treatments was chosen for the field experiment. The chlorfenvinphos treatment at 22 g a.i./unit was also included, as the higher dose had reduced plant vigour.

Table 2. The percentage germination of untreated turnip seed and turnip seed treated with three doses of chlorpyrifos, chlorfenvinphos, fipronil and spinosad.

Insecticide	Actual dose (g a.i./unit)	Germination (% ± se) 11 days	19 days
Chlorfenvinphos	11	90 ± 1.7	92 ± 1.2
	22	89 ± 1.3	93 ± 1.4
	50	71 ± 2.6	81 ± 2.0
Chlorpyrifos (Empire)	8	94 ± 1.4	94 ± 1.3
	14	92 ± 1.7	94 ± 1.1
	32	97 ± 1.0	98 ± 0.6
Chlorpyrifos (Gigant)	8	89 ± 1.8	92 ± 1.5
	18	94 ± 1.2	95 ± 1.2
	40	94 ± 1.3	95 ± 1.2
Spinosad	38	93 ± 1.3	94 ± 1.1
	79	94 ± 1.5	97 ± 1.2
	179	94 ± 1.2	97 ± 1.0
Fipronil	24	94 ± 0.9	95 ± 0.8
	48	95 ± 1.1	97 ± 0.7
	80	94 ± 1.4	96 ± 1.1
Untreated	0	94 ± 1.4	95 ± 1.2

b) Assessing the seed treatments against a field population of cabbage root fly.

Materials and methods

An area of eight (1.83 m wide x 20 m long) seedbeds was prepared in the experimental area at HRI Wellesbourne. Each of the inner six beds was divided to give six 2.5 m plots, with 1 m between each plot. On 28 June, before the start of egg laying by the second fly generation, the turnip plants (from the previous glasshouse trial) were transplanted, at 50 cm spacing, into a field plot. The five insecticide treatments and the untreated plants were arranged in a 6 x 6 plot block. Each plot was planted with twenty four (6 x 4) plants of one insecticide treatment. The chlorfenvinphos treated plots were planted with twelve (6 x 2) plants of each test dose.

From 3-9 September, 15 cm diameter x 15 cm deep soil cores were taken from around the roots of 6 plants in each sub-plot and all of the roots were harvested. Cabbage root fly pupae were extracted from the soil samples by flotation in water and the numbers of pupae recovered were counted. The roots were washed, weighed and scored for cabbage root fly damage. The root damage index was calculated based on scoring root damage from 0 (no damage) to 4 (>50% damage). The mean numbers of cabbage root flies recovered from the soil samples and the mean root damage index were subjected to Analysis of Variance. The insect counts were square root transformed prior to analysis.

<u>Results</u>

Egg laying by the second generation of cabbage root flies started to increase from mid-July onwards and peak numbers of eggs were laid at the end of July (egg samples taken from a nearby monitoring plot).

The chlorfenvinphos (50 g a.i./unit) treatment reduced the numbers of larvae and pupae recovered compared with the untreated control (p=0.05) (Figure 1), but only small differences in the levels of root damage were observed compared with the untreated control (Figure 2). The higher rate (50 g a.i./unit) chlorfenvinphos treatment would not be agronomically acceptable, due to the problems of phytotoxicity. There was evidence of predation on cabbage root fly eggs and larvae in the untreated plots, as more pupae were recovered from the roots treated with chlorpyrifos and fipronil. Without this factor, the damage to untreated roots may have been far greater.

Figure 1. The mean numbers of larvae and pupae recovered from turnip roots treated with insecticidal seed treatments.

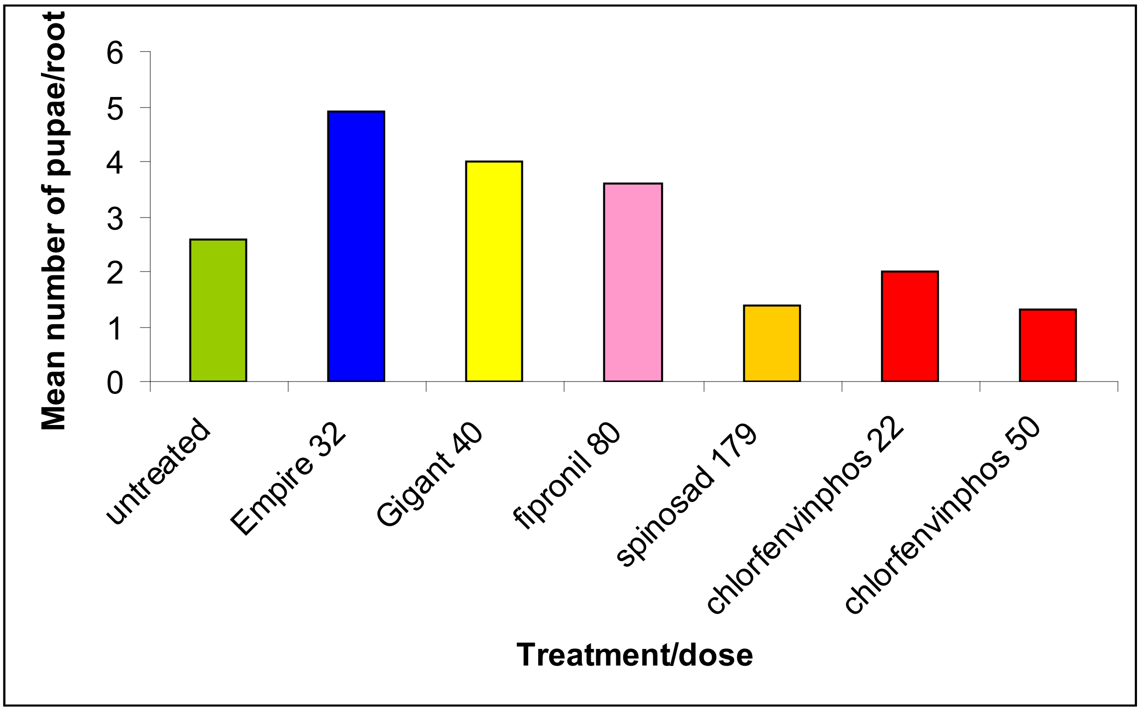
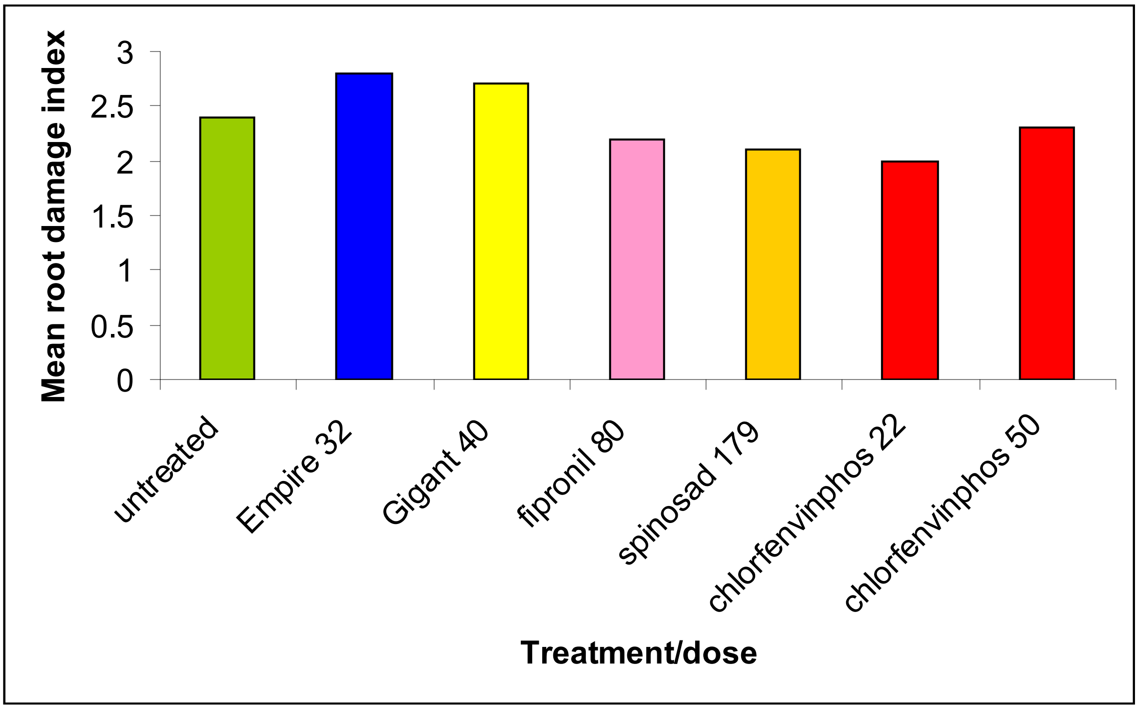


Figure 2. The mean root damage index of turnip roots treated with insecticidal seed treatments.



B. Insecticide treated bait

Experiment 2. Will any of the currently available insecticides kill adult flies if incorporated into one of the commercially available insect baits?

The only commercially available treated bait that could be obtained was GF 120 Naturalyte Fruit Fly Bait (Dow Agrosciences), which contained spinosad (0.02% a.i.). The product is designed to kill fruit flies and is not available in the UK. At the manufacturer's recommended dilution rate, the final spinosad concentration is 80 mg a.i./l. This concentration was used as a basis for all of the tests conducted.

a) Insecticide screening technique

Materials and methods

Due to the temporary unavailability of laboratory-reared cabbage root flies at the start of these experiments, initial insecticide screening was conducted against a closely related species – the onion fly (*Delia antiqua*).

Insecticide bait solutions were prepared to give insecticide concentrations of 80 mg/l. The bait consisted of a solution of sucrose (10%) and yeast extract (1%) with one of the test insecticides. The insecticides tested were chlorpyrifos (Dursban), fipronil (50% a.i. ST formulation), imidacloprid (Gaucho), lambda-cyhalothrin (Hallmark), pirimicarb (Aphox) and spinosad (Spintor). Adsorbent cotton wool was placed into open petri dishes and fully saturated (40 ml) with one of the bait solutions. Samples of untreated sucrose (10%) solution were also retained on cotton wool in petri dishes.

Fifteen male and fifteen female onion flies (5-7 days old) were placed into nylon net cages (35 cm square) and transferred to an illuminated glasshouse (set to give a 16-hour day length). One insecticide-treated dish was placed into each cage. Two dishes of each treatment were tested, one with, and one without, an additional dish of untreated sucrose solution (choice of food). Additionally, a bottle containing fresh water and a blotting paper wick was placed into each cage. One cage contained only untreated sucrose solution and one cage contained no food source. The numbers of dead flies of each sex were recorded twice a day for the following 3 days.

Results

During the course of the trial, just 1 fly (3% of all the male and female flies tested), which had only untreated sucrose as a food source, died. A total of 60% of males and 20% of females had died after 3 days when given no food at all. All of the insecticide-treated baits killed flies (Table 3). It is not clear whether all of the flies were killed by the ingestion of insecticide, or if some were killed indirectly by the insecticides acting as feeding deterrents. When the flies were offered both untreated and treated food, mortality was lower with all of the insecticides tested. The male flies (which are smaller) generally died sooner than the female flies, but this was not always the case.

Table 3. The cumulative percentage mortality of onion flies which died after feeding on insecticide treated bait solutions.

		Days after start of experiment					
	± untreated	Male flies			Female flies		
Treatment	sucrose	1	2	3	1	2	3
Untreated	+	0	0	0	7	7	7
No food	-	27	60	60	20	20	20
Chlorpyrifos	+	40	73	80	47	80	87
Chlorpyrifos	-	72	89	100	50	93	100
Fipronil	+	33	73	80	80	100	100
Fipronil	-	87	100	100	93	93	93
Imidacloprid	+	47	67	73	7	20	33
Imidacloprid	-	73	87	93	13	80	87
Lambda-cyhalothrin	+	13	53	53	13	53	60
Lambda-cyhalothrin	-	67	87	93	33	67	67
Pirimicarb	+	47	60	60	13	20	27
Pirimicarb	-	67	73	73	20	53	60
Spinosad	+	47	60	73	80	93	100
Spinosad	-	78	94	100	85	100	100

b) Composition of bait

The insecticide screening experiment confirmed that a solution containing sucrose (10%) and yeast extract (1%) was an attractive food source for the onion fly. The commercial bait (GF120) contains sugars, vegetable proteins and propylene glycol in unspecified proportions. The propylene glycol keeps the bait in a liquid form after spraying and therefore keeps it more readily accessible to the flies.

Materials and methods
A range of solutions containing varying concentrations of sucrose, yeast extract and propylene glycol were prepared and compared for ease of preparation and the longevity of the droplets. As brassicas have waxy leaves, problems with adhesion to the leaf surface were anticipated. To test adhesion, droplets (20 µl) of the test solutions were applied to cauliflower leaves using a Gilson pipette. Varying amounts of Codacide oil (Microcide) were added to aid adhesion.

Results
All results of all tests are indicative only. The sucrose became difficult to dissolve above concentrations of around 20%. The droplets that kept their form the longest contained 20% sucrose, 5% yeast extract and 10% propylene glycol. None of the solutions tested adhered very well to cauliflower leaves without added codacide. The optimum codacide concentration appeared to be about 0.5%. At this level, adhesion to the cauliflower leaves was improved. However, increasing the concentration to 1% caused excessive spreading of the droplets.

c) Application method

Materials and Methods
Dow Agrosciences recommend a droplet size of 4-6 mm for their GF-120 Naturalyte Fruit Fly Bait. This equates to a droplet volume of about 20 µl. Preliminary trials with the GF-120 and the other laboratory prepared baits highlighted the difficulty of applying large droplets to waxy brassica leaves. Hence an alternative method was investigated.

The GF-120 bait concentrate was diluted (1:1.5) with water to give a final spinosad concentration of 80 mg/l. An “HRI” bait was prepared by dissolving sucrose (20% w/v) and yeast extract (5% w/v) in water:propylene glycol (9:1) and adding codacide (0.5% v/v). Spinosad (Spintor) was added to give a concentration of 80 mg a.i./l. A portion of the bait was left untreated.

Glasshouse-grown cauliflowers were treated with the baits using a Gilson pipette (20 µl, 8 droplets/plant) or hand sprayer (approximately 5 ml/plant). The GF-120 bait was applied using only the former method and the “HRI” bait was applied using both methods. Untreated control plants were prepared by spraying with untreated “HRI” bait and treated control plants were prepared by spraying with spinosad only (80 mg a.i./l).

Fifteen male and fifteen female cabbage root flies (5-7 days old) were placed into nylon net cages (35 cm square) and transferred to an illuminated glasshouse (set to give a 16-hour day length). One treated plant was placed into each cage. Two plants of each treatment were tested, one with, and one without, an additional dish of untreated sucrose solution (choice of food). Additionally, a jar containing fresh water and a blotting paper wick was placed into each cage. The numbers of dead flies of each sex were recorded twice a day for the following 3 days. The experiment was repeated on two further occasions.

Results

Fly mortality is summarised in Table 4. Spinosad without bait did not increase mortality compared with the untreated control, so it can be inferred that this insecticide has to be ingested to kill cabbage root flies. The commercial, GF-120, bait performed poorly, but it should be noted that it was developed to kill fruit flies. When there was no other food source available, it killed only male flies and when there was an alternative food source, no flies (above natural mortality) were killed. The "HRI" bait was more effective either as droplets or as a spray. The spray performed better than the droplets and there was only a small reduction in efficacy when an alternative food source was available.

Table 4. The mean cumulative percentage mortality of cabbage root flies which died after feeding on spinosad-treated bait solutions applied as large droplets or using a hand sprayer.

		Days after start of experiment					
	± untreated	Male flies			Female flies		
Treatment	sucrose	1	2	3	1	2	3
Untreated	+	0	11	14	0	4	15
Spinosad only	+	0	4	4	0	0	7
GF-120	+	0	0	8	0	0	7
GF-120	-	7	21	50	0	0	8
"HRI" droplet	+	2	42	66	6	27	58
"HRI" droplet	-	12	53	82	0	26	68
"HRI" spray	+	7	59	82	5	72	90
"HRI" spray	-	24	83	93	8	62	86

Based on this information, it was decided to conduct future experiments using the spray technique, as it was easier to apply, more stable and more effective. All further experiments also included an alternative, untreated, food source to mirror field conditions where there would be a number of alternative food sources available.

d) The effect of insecticide and dose on the mortality of adult cabbage root flies

Materials and methods

On 15 November, one 308 Hassy tray was sown with cauliflower seed. When the seedlings had reached the 4-leaf stage they were transplanted into Optipot 11M pots containing Levington compost. These plants were used for all the experiments.

Five insecticides were tested: chlorpyrifos (organophosphate), lambda-cyhalothrin (pyrethroid), pirimicarb (carbamate), fipronil and spinosad (spinosyn), representing a range of insecticide chemistries. The doses tested were 20 mg a.i./l, 80 mg a.i./l (GF-120 dose) and 320 mg a.i./l. The "HRI" bait was used and all treatments were sprayed onto cauliflower plants. From 10 March to 11 April a series of trials were set-up in a glasshouse. Each trial consisted of one plant treated with each insecticide at a single dose and an untreated control plant sprayed with insecticide-free “HRI” bait. Fresh bait was prepared for each trial and each insecticide dose was tested on three occasions.

Twenty five male and twenty five female cabbage root flies (1-5 days old) were placed into nylon net cages (35 cm square) and transferred to an illuminated glasshouse (set to give a 16-hour day length). One treated plant was placed into each cage. In addition, a dish containing sucrose solution retained on cotton wool and a bottle containing fresh water and a blotting paper wick were placed into each cage. The numbers of dead flies of each sex were recorded twice a day for the following 3 days.

Results

Cabbage root fly mortality recorded over the 3 days after treatment is shown in Figure 3 (20 mg a.i./l), Figure 4 (80 mg a.i./l) and Figure 5 (320 mg a.i./l). The mortality of cabbage root flies was consistently low in the untreated control cages in all of the trials. As seen previously, male flies generally died more quickly than female flies, irrespective of treatment.

Pirimicarb was virtually non-toxic to the flies at all of the doses tested, despite showing activity against the onion fly. Fipronil was consistently the most effective insecticide at all of the test doses against both sexes. Lambda-cyhalothrin, chlorpyrifos and spinosad performed similarly against male flies at 320 mg a.i./l, but generally lambda-cyhalothrin was less effective than chlorpyrifos, which was less effective than spinosad. At the lower dose (20 mg a.i./l), lambda-cyhalothrin and chlorpyrifos had little effect.

Figure 3. The mean cumulative mortality of cabbage root flies exposed to insecticide treated bait at a concentration of 20 mg a.i./l.

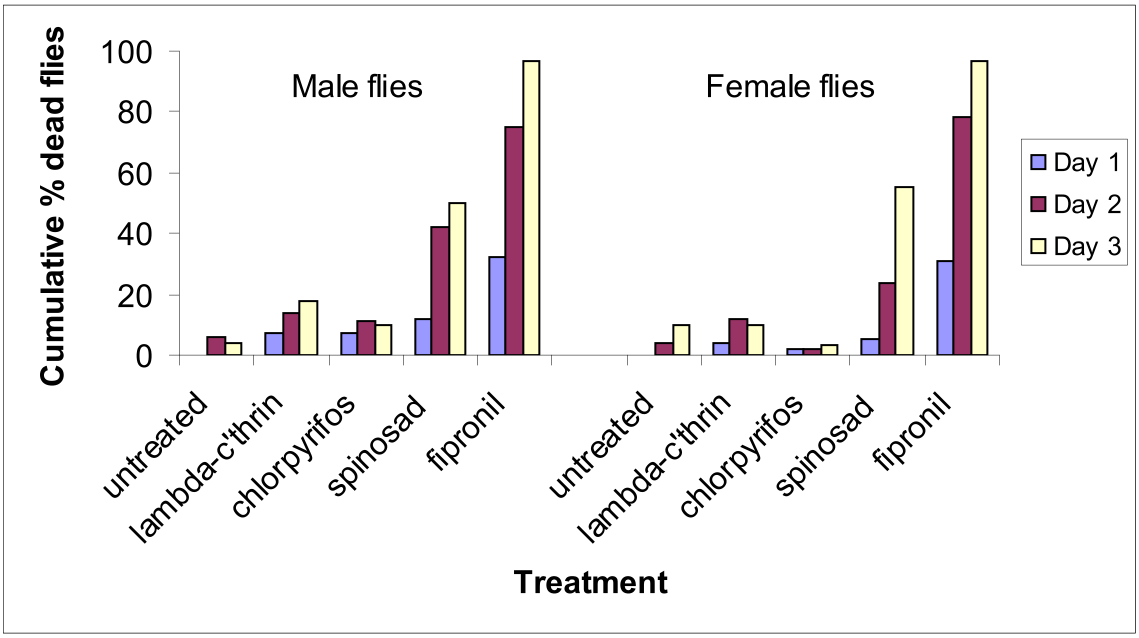


Figure 4. The mean cumulative mortality of cabbage root flies exposed to insecticide treated bait at a concentration of 80 mg a.i./l.

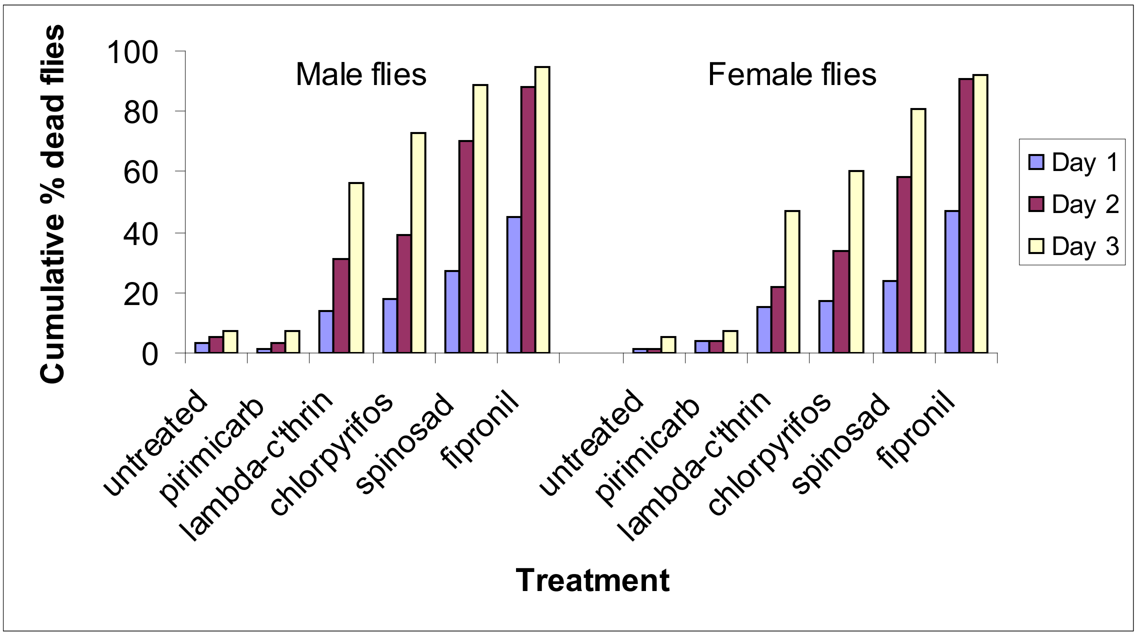
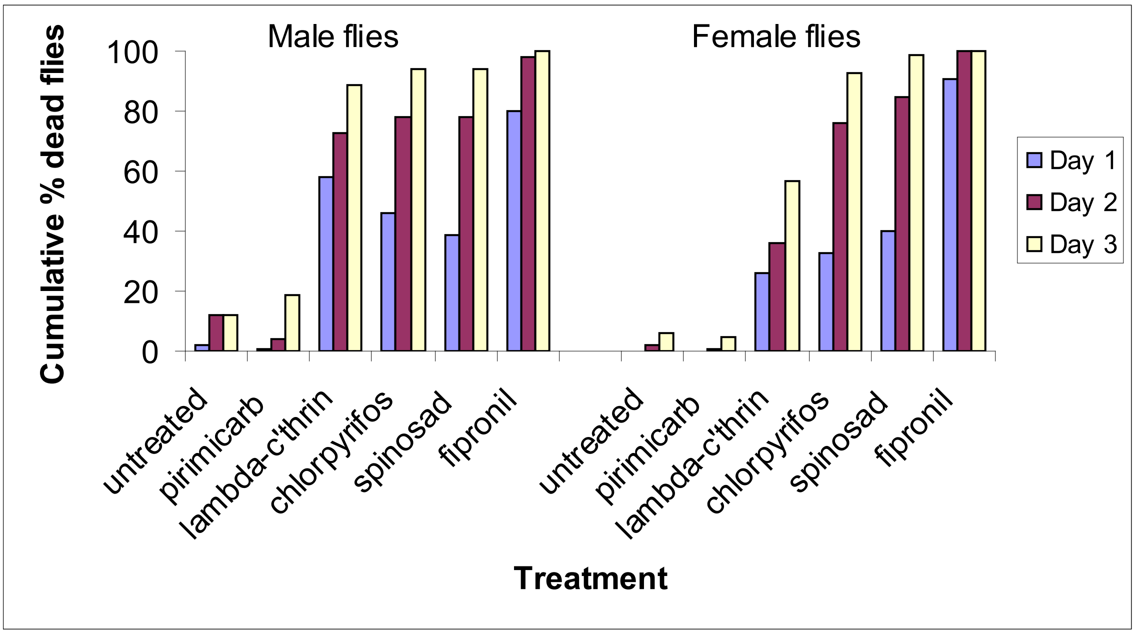


Figure 5. The mean cumulative mortality of cabbage root flies exposed to insecticide treated bait at a concentration of 320 mg a.i./l.



e) The effect of aged residues on mortality of adult cabbage root flies

<u>Materials and methods</u>

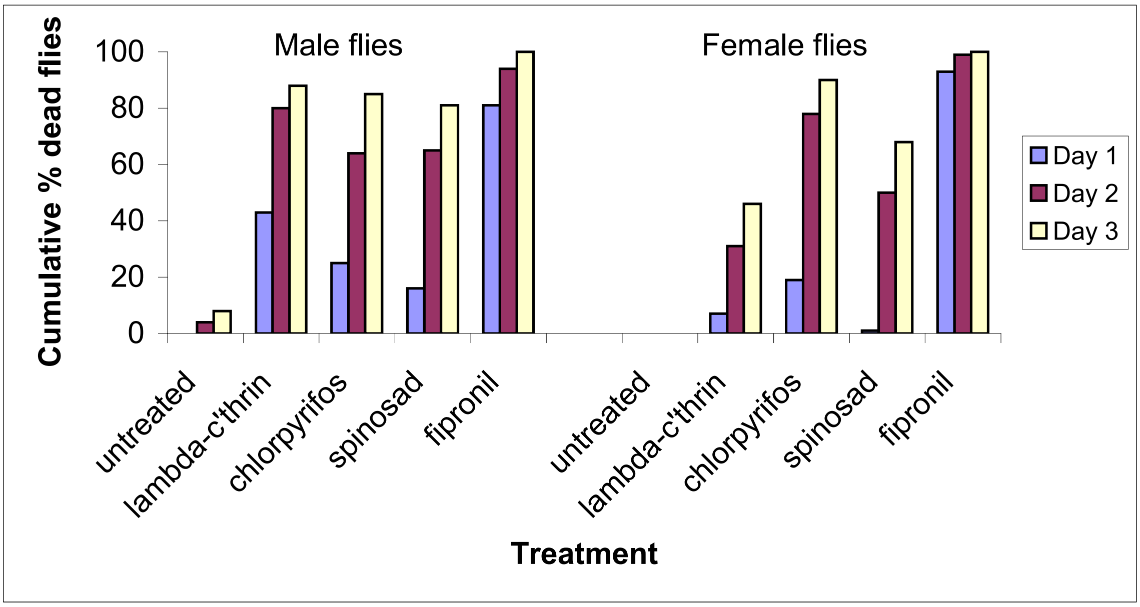
On 2 April, lambda-cyhalothrin, chlorpyrifos, spinosad and fipronil treated baits were prepared as described previously. Three cauliflowers were sprayed with each of the bait solutions using a hand sprayer. The plants were kept under glasshouse conditions. Extra care was taken to ensure

that the treated plants did not touch each other and thereby transfer bait from one plant to another. On 16 April (2 weeks after treatment), twenty five male and twenty five female cabbage root flies (1-5 days old) were placed into nylon net cages (35cm square) and transferred to an illuminated glasshouse (set to give a 16-hour day length). One of the previously treated plants was placed into each cage. In addition, a dish containing sucrose solution retained on cotton wool and a bottle containing fresh water and a blotting paper wick was placed into each cage. The numbers of dead flies of each sex were recorded twice a day for the following 3 days.

<u>Results</u>

The results obtained using aged residues (Figure 6) cannot be compared directly with those obtained using fresh residues. However, it is clear that all four insecticide baits were attractive to the flies, and toxic. Fipronil lost little or no killing power at this dose even two weeks after treatment. The three other insecticides appeared to be slightly less effective than they were as freshly applied residues. Spinosad was clearly less active than chlorpyrifos against female flies when, with fresh residues, there was little difference between the two insecticides. Of the four test insecticides, spinosad is inherently the least stable, so it can be assumed that a certain amount of chemical breakdown had occurred.

Figure 6. The mean cumulative mortality of cabbage root flies exposed to insecticide treated bait at a concentration of 320 mg a.i./l, which had been previously aged for 2 weeks in a glasshouse.



C. Insect deterrents

Experiment 3. How effective under laboratory and field conditions are five products (Antistress, Ecoguard, Ecospray, Majestik and Seagrow) which, it has been suggested, are capable of deterring the cabbage root fly from laying on brassica plants?

a) Laboratory trials

Materials and methods

On 2 September, one 308 Hassy tray was sown with swede seed. When the seedlings had reached the 4-leaf stage, they were transplanted into Optipot 9M pots containing Levington compost.

1) Laboratory "choice" experiments

To ensure that all the plants were exposed equally to the test insects, a rotating cage was used. The cage consisted of two sealed and illuminated chambers, one on top of the other. The cage lights were set to give a 16-hour light period. Each chamber contained a circular turntable that could be rotated at a constant speed. Four removable segments (to support the test plants) were placed onto each turntable. These covered the four quadrants of the turntable exactly.

Because the experiments were replicated in time, cabbage root flies of a known age were used on each test occasion, so that the results of different tests could be compared. One potted swede plant was placed into each segment and freshly sieved soil was applied until the whole segment was covered, to provide a bare soil background. Two treated and two untreated plants were placed into each compartment, so that similar treatments were located opposite one other. Ecospray (Ecospray Limited), Antistress (Agrichandlers) and Majestik (Hortichem) treatments were sprayed onto the plants before they were placed in the segments. Ecoguard (Ecospray Limited) and Seagrow (A. B. Bell & Sons) treatments were placed onto the soil surface around the base of each plant once the pots had been set up in the segments. The treatment rates are shown in Table 5.

Twenty, six-day old, gravid female cabbage root flies were introduced into each cage containing the test plants and allowed to lay eggs for 24 hours. The plants were then carefully removed from the segments and the surrounding soil was washed off into plastic dishes. The dishes were filled with water and the soil was agitated by stirring. The soil was allowed to settle for about 1 hour and the cabbage root fly eggs, which float on the surface of the water, were counted. Each treatment was repeated on five occasions. The results were subjected to a GLM analysis with binomial errors and logit link to determine whether the proportion of eggs laid on the treated plants was affected by the treatment applied.

Table 5. The doses of proposed cabbage root fly repellents applied to swede plants in laboratory "choice" and "no choice" experiments.

Treatment	Dose	Application method
Antistress	50 ml/l	spray to run-off
Ecospray	20 ml/l	spray to run-off
Majestik	50 ml/l	spray to run-off
Ecoguard	0.25 g/plant	placed around stem base
Seagrow	16 g/plant	placed around stem base

2) Glasshouse "no choice" experiments

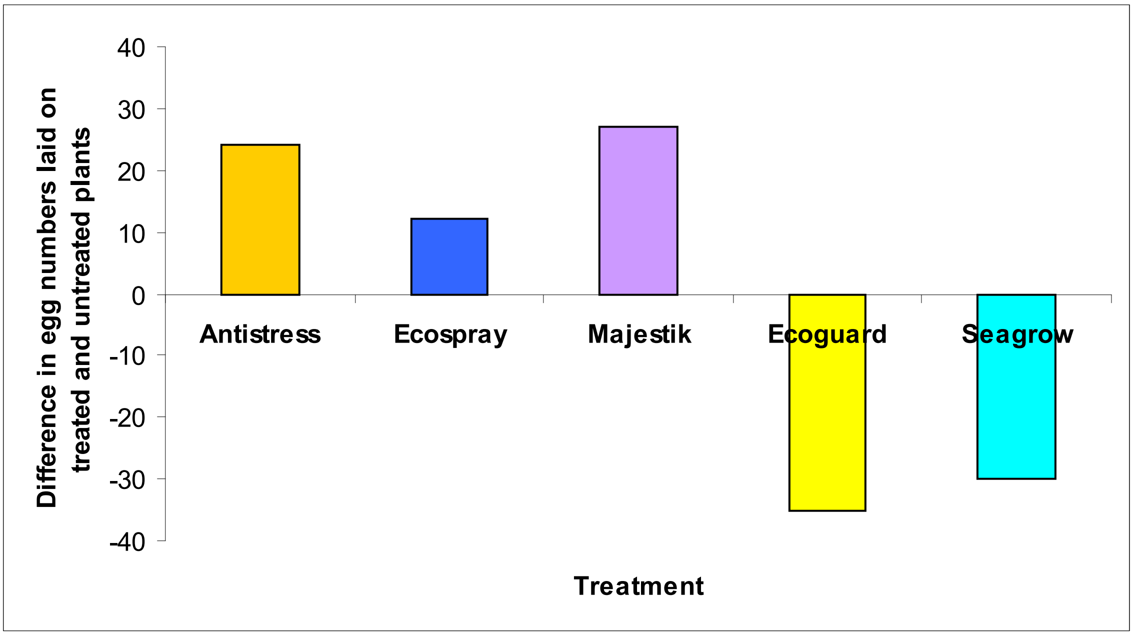
With the "no choice" experiments, a rotating cage was unnecessary, so the experiments were conducted in a glasshouse (20 ± 2ºC). The glasshouse lights were set to give a 16-hour light period. Sets of four swede plants were sprayed with Antistress, Ecospray or Majestik as in the "choice" experiment. Each of the three sets of treated plants was placed in a tray under a separate clear plastic cage (100cm wide x 50cm deep x 60cm tall). A further three cages contained untreated plants. All the cages (6 in total) were placed on a bench in the glasshouse. The surface of the pots and the surrounding area were then covered with sieved soil to provide a bare soil background. Finally, the Ecoguard and Seagrow treatments were applied to the soil in two of the three cages containing untreated plants, whilst the soil in the remaining cage was left untreated. Twenty, six-day old, gravid female cabbage root flies were introduced into each cage and the plants were removed after 24 hours. The cabbage root fly eggs were extracted and counted as described earlier. The results were subjected to GLM analysis assuming a Poisson distribution and log link to assess the numbers of eggs laid on treated plants relative to the untreated control.

Results

1) "Choice" experiments

The differences between the numbers of eggs laid on treated and untreated plants are displayed in Figure 7. When the female cabbage root flies were offered a choice of treated or untreated plants, both of the treatments that had been applied to the soil surface (Ecoguard and Seagrow) appeared to deter the flies, to some extent, from laying eggs. In particular, a smaller proportion of eggs was laid on plants treated with Ecoguard or Seagrow than on plants treated with Antistress or Majestik ($p<0.01$). However, the mean numbers of eggs recovered from the Ecoguard and Seagrow treatments still exceeded 30 per plant.

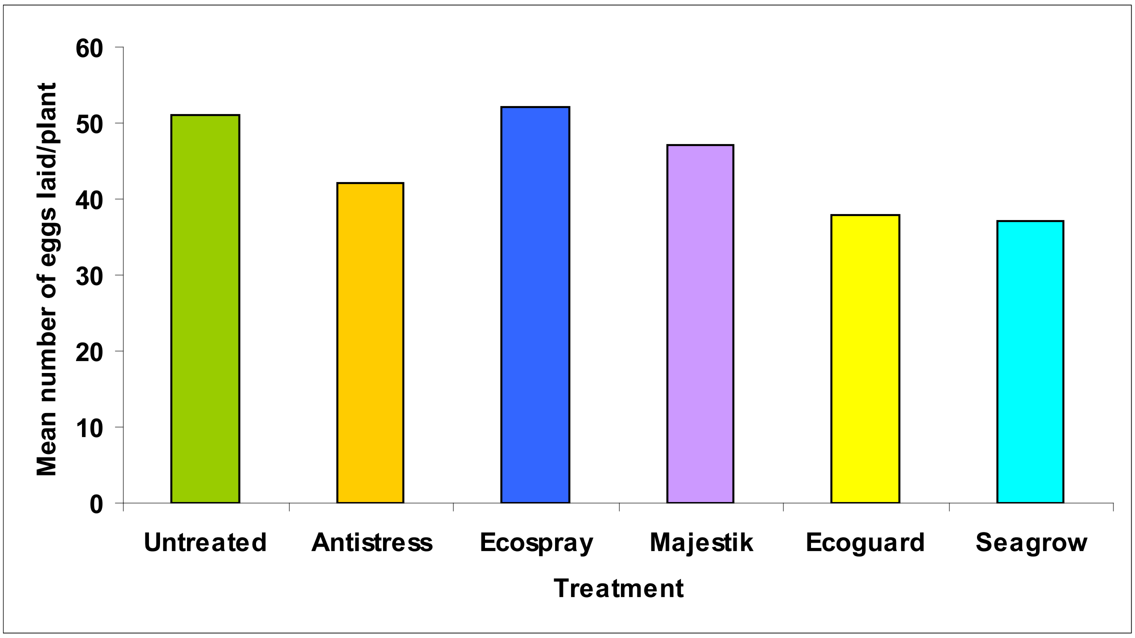
Figure 7. The mean difference between treated and untreated swede plants in the numbers of cabbage root fly eggs laid in "choice" experiments.



2) "No choice" experiments.

The mean numbers of eggs laid on plants treated with the five treatments, or left untreated, are displayed in Figure 8. When the female cabbage root flies were presented with plants treated similarly, their only choice was to lay or not to lay. As in the "choice" experiments, both Ecoguard and Seagrow appeared to reduce egg laying and Antistress had a similar effect. However, reductions in egg numbers did not exceed 30% compared with the untreated control, and the mean numbers of eggs laid on all of the treated plants still exceeded 35 per plant. None of treatments produced a statistically significant reduction in egg numbers compared with the untreated control.

Figure 8. The mean numbers of cabbage root fly eggs laid on treated and untreated swede plants in "no choice" experiments.



b) Field experiment

<u>Materials and Methods</u>

Three 308 Hassy trays, sown with untreated swede seeds on 24 May, were maintained under glasshouse conditions. On 21 July, the swede plants were transplanted, at 20 cm spacing, into a field plot, which was made up of 3 blocks of 6 plots. Each plot contained 25 plants and was separated from adjacent plots by 1 m of bare soil. The plants were covered immediately with Envirofleece (Agralan) to keep out any first generation cabbage root flies.

On 23 July, prior to peak egg laying by the second generation of the cabbage root fly, the fleece was removed. One plot in each block was treated with Antistress, Antistress + Garlick (garlic extract), Majestik, Ecoguard, Seagrow or left untreated. The treatments were fully randomized. The doses and method of application are shown in Table 6. To maximise any potential effects, Majestik (which could have been washed off by rainfall) and Seagrow were re-applied at weekly intervals on a further four occasions.

On 30-31 August, 15 cm diameter x 15 cm deep soil cores were taken from around the roots of 6 plants in each plot. Cabbage root fly pupae were extracted from the soil samples by flotation in water and the numbers of fly pupae were counted. The remaining roots were harvested and the foliage discarded. After washing, the roots were assessed for cabbage root fly feeding damage. The damage was scored on a scale of 0 (no damage) – 4 (>50% damage). The mean numbers of insects recovered and the mean root damage index were subjected to Analysis of Variance. The insect data were square root transformed prior to analysis.

Table 6. The dose and method of application of deterrent products applied to a field trial.

Product	Dose	Application method
Antistress	50ml/l	Spray to run-off with knapsack sprayer
Antistress + Garlick	50ml/l + 10ml/l	Spray to run-off with knapsack sprayer
Majestik	25ml/l	Spray to run-off with knapsack sprayer
Ecoguard	16kg/ha	Applied around root at 0.6g/m row
Seagrow	410g/m^2	Applied around root

<u>Results</u>

Root damage due to feeding by cabbage root fly larvae was heavy on all plots. None of the treatments reduced the numbers of cabbage root fly larvae and pupae recovered (Figure 9) or the amount of root damage (Figure 10). Despite showing evidence of some repellent effect in laboratory trials, Seagrow significantly increased (p=0.05) the level of damage relative to the untreated plots. This may be because it improved the conditions for larval survival by acting as mulch, thereby retaining more moisture in the soil around the root.

Figure 9. The mean numbers of cabbage root fly larvae and pupae recovered from around swede roots after treatment with insect deterrents.

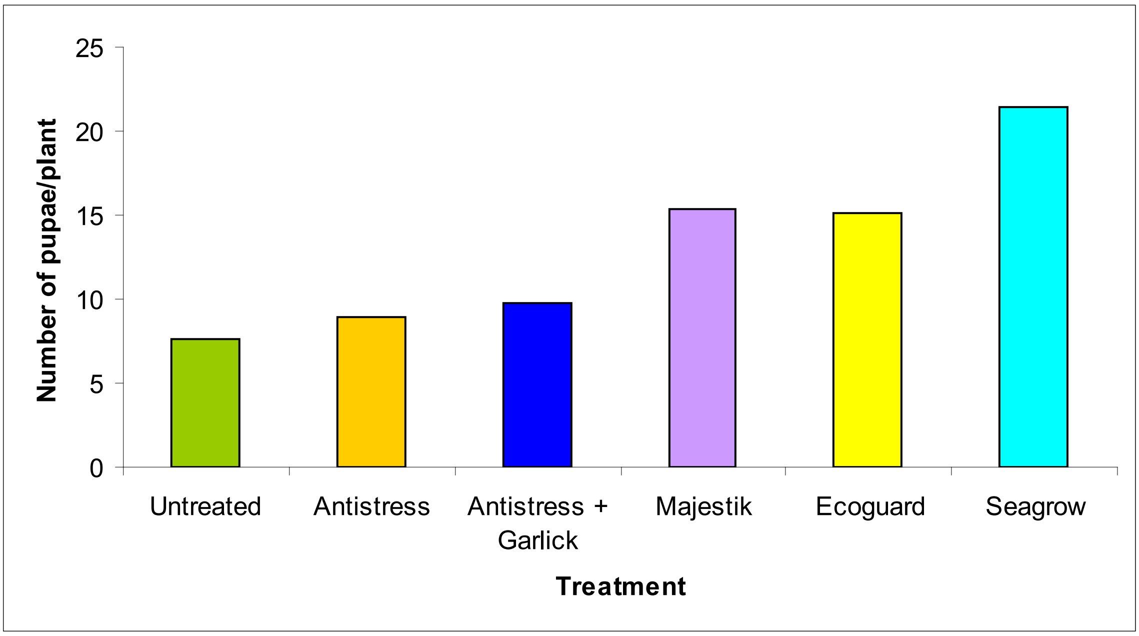
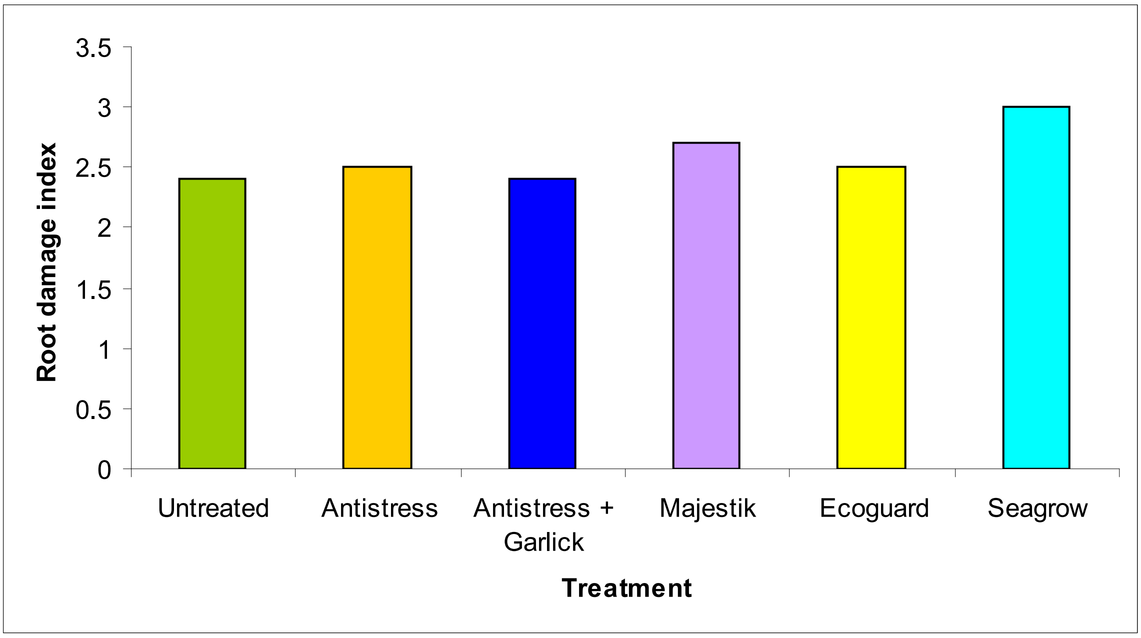


Figure 10. The mean root damage index of swede roots after treatment with insect deterrents.



CONCLUSIONS

Film-coated seed

Contrary to expectation, relatively high doses (40g a.i./100,000 seeds) of chlorpyrifos (Gigant or Empire formulations) were not phytotoxic to turnip seeds. This is because the chlorpyrifos was stabilised on Talcum powder. In contrast, chlorfenvinphos showed signs of phytotoxicity at lower doses (20g a.i./100,000 seeds).

In the field trial, the efficacy of all the insecticide treatments tested was disappointing. In the case of the chlorpyrifos and fipronil treatments, the numbers of cabbage root fly pupae and the levels of root damage actually increased compared with untreated controls. Both treatments had previously shown a measure of cabbage root fly control (Jukes *et al.,* 2000, 2001) so it is assumed that there was a high level of predation on cabbage root fly eggs and larvae in the untreated plots, but that the predators were killed in the treated plots. Spinosad and chlorfenvinphos were both partially effective. The relative effectiveness of spinosad is all the more interesting because it is almost certainly the most readily degraded chemical of all those that were tested.

Insecticide treated baits

Glasshouse trials with insecticide-treated baits showed that it is possible to kill adult onion flies and cabbage root flies using this method of application. They also confirmed that insecticides have to be ingested to be effective against adult flies. A total of six insecticides with varying chemistries were tested and all six killed onion fly adults. In trials with the cabbage root fly, pirimicarb was ineffective. Of the other four insecticides tested, fipronil was consistently the most potent. Chlorpyrifos and spinosad performed similarly, but spinosad residues were not as persistent. Lambda-cyhalothrin generally killed the fewest flies. Imidacloprid was not tested against the cabbage root fly.

Observations during the trials indicated that the flies spent very little time feeding and thus death might often have been the result of a single feeding event. At the highest and most effective dose tested (320 mg a.i./l), a field application at standard spray rates (300l/ha) would equate to about 100 g a.i./ha, but adequate leaf coverage could probably be achieved at considerably lower spray rates.

Despite the obvious potential of baits there are a number of unanswered questions:

1) Will the treatments work under field conditions?
2) Is the crop foliage the most appropriate carrier for baits?
3) Will insecticide stability become a problem under field conditions?
4) Is there the potential to improve the composition of the baits (there could be a compromise between ensuring baits are attractive, but not making them so nutritious that only small amounts are imbibed)?
5) Are there more effective chemicals that are available currently and which are more toxic?
6) Will female flies lay eggs between ingesting insecticide and dying?

<u>Insect deterrents</u>
The potential deterrents performed poorly under field conditions. Although the two soil-applied products, Ecoguard (garlic granules) and Seagrow (composted seaweed), showed some deterrent effect in laboratory trials, they had no effect when used in the field. In fact rather than reducing the numbers of cabbage root fly pupae, Seagrow increased them. Although Seagrow may well have had some deterrent effect, it also appeared to provide ideal soil conditions for the survival of fly larvae. The foliar treatments (Antistress, Majestik and Ecospray) showed little effect as deterrents. It can be assumed that their odour and/or physical properties were not sufficient to mask the characteristics of the swede leaves beneath them.

TECHNOLOGY TRANSFER

14-Aug-01	Field experiments visited by members of Brassica Growers Association.
16-Jan-02	Brassica Conference – talk by Stan Finch on cabbage root fly control.

GLOSSARY

a.i.	active ingredient
mg	milligram or one-thousandth of a gram (g)
OP	organophosphorus
PVA	polyvinyl acetate – sticks the insecticide onto the seed coat
Unit	100,000 seeds

ACKNOWLEDGEMENTS

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