## FINAL REPORT

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Brassica crops: Evaluation of non-organophosphorus insecticides for controlling the cabbage root fly

FV 242a

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## **Grower Summary**

## FV 242a

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Final report 2004

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## FV 242a

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

## Headline

- Note The only product currently approved (May 2004) for CRF control on swede and turnip is chlorpyrifos seed treatment (on imported seed).
- When applied as a seed treatment to swede at a sufficiently high dose, spinosad provided control of first generation cabbage root fly larvae and partial control of the second generation. However, the dose recommended currently by the manufacturers of spinosad may not be sufficiently high to provide economic levels of control. By inference, the results suggest that the seed treatment alone would provide adequate protection for module raised leafy brassicas, where lower levels of control are required.
- Although freshly applied insecticide + bait solutions were effective against adult cabbage root flies and killed female flies before they laid eggs, foliar spray treatments did not persist long enough to provide effective control of field populations of cabbage root fly.
- Drench treatments with thiacloprid or teflubenzuron and seed treatments with thiamethoxam or cyromazine all controlled cabbage root fly and, with the exception of teflubenzuron, there was a pronounced dose response effect. These results confirm that apart from spinosad, several active ingredients appear to work effectively as soil insecticides for cabbage root fly control and have the potential to be used as drench or seed treatments on leafy brassicas.
- Although fresh garlic granules controlled cabbage root fly larvae, their efficacy declined as the granules aged.
- Carbosulfan does not have the same efficacy against cabbage root fly larvae as carbofuran. In the single dose trial conducted in this project there was some evidence of a reduction in the numbers of cabbage root fly larvae present, but this was insufficient to reduce root damage. However, carbosulfan could be an effective treatment at drilling for the control of flea beetles and aphids.
- Although there are a number of potential treatments for control of cabbage root fly on leafy brassicas, no completely effective method of controlling cabbage root fly on established crops of swede has been identified. The key question is 'how can the soil-active insecticides identified be applied so that they are in the right location to protect long-season crops such as swede'?

### **Background and expected deliverables**

Brassica crops are grown currently on approximately 30,000 ha in the UK and the marketed value of these crops is about £150M/annum [*Basic Horticultural Statistics for the United Kingdom. Calendar and Crop Years 1991/92 – 2001/02. Department for Environment, Food and Rural Affairs, National Statistics*]. 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, as a result of the UK/EU pesticide reviews,

some products have been withdrawn already and others may be withdrawn in the future. There are now only two approved chemicals; carbosulfan (Marshall) and chlorpyrifos (Dursban), for cabbage root fly control on leafy brassica crops in the UK. Since 31 December 2003, no product has been available to control the cabbage root fly on swede and turnip, 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. Owing to the concern being expressed by swede growers, the experiments in this project concentrated on swede crops. 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.

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. The expected deliverables from this work include:

- An evaluation of the persistence of spinosad applied as a seed treatment for control of first and second generation cabbage root fly.
- An indication of the field performance of foliar sprays of insecticide + sugar and yeast extract baits against cabbage root fly.
- An assessment of the activity and persistence of insecticide + bait treatments.
- An indication of the effectiveness of the novel insecticides teflubenzuron, thiacloprid, cyromazine and thiamethoxam against cabbage root fly when applied as drench or seed treatments.
- An indication of the persistence of garlic granules applied to control cabbage root fly eggs and larvae.
- An evaluation of the performance of carbosulfan granules applied to swede plots at drilling to control cabbage root fly on swede.

#### Summary of the project and main conclusions

Seven experiments were done in 2003-4 using eight insecticides (Tracer (spinosad), Posse (carbosulfan), Dursban (chlorpyrifos), Dimethoate 40 (dimethoate), Triguard (cyromazine), Nemolt (teflubenzuron), Calypso (thiacloprid), Actara (thiamethoxam)). In addition, a potential 'natural' insecticide (ECOguard granules) was assessed.

Experiments were done to answer the following questions:

- 1. How long do spinosad seed treatments persist?
- 2. Will weekly spray applications of spinosad + bait control adult cabbage root flies in the field?
- 3. Can the effectiveness of insecticide baits be improved and do female cabbage root flies that are exposed to insecticide baits lay eggs before they die?
- 4. How does exposure to natural daylight for different periods affect the effectiveness of insecticide + bait mixtures?

- 5. How effective are the novel insecticides thiamethoxam, cyromazine, thiacloprid and teflubenzuron against cabbage root fly when applied as drench or seed treatments?
- 6. How long does the efficacy of Ecoguard garlic granules persist?
- 7. Will carbosulfan granules applied at drilling control cabbage root fly larvae on swedes?

#### Main conclusions

- When applied as a seed treatment to swede at 72 or 100 g a.i./100,000 seeds, spinosad provided partial control of two generations of cabbage root fly larvae and reduced seedling losses.
- When applied in a bait solution, fresh applications of spinosad, chlorpyrifos and dimethoate were effective at killing caged adult cabbage root flies. After exposure to natural daylight the efficacy of these three treatments diminished rapidly.
- The composition of the bait used to kill adult cabbage root flies can be varied considerably without reducing efficacy.
- When applied in a bait solution and sprayed weekly for the duration of the second generation of the fly, spinosad and chlorpyrifos were not effective at controlling cabbage root flies in the field.
- In glasshouse trials, teflubenzuron applied as a drench to plant modules was highly effective at controlling cabbage root fly eggs/larvae. Thiacloprid applied similarly was less effective.
- In glasshouse trials, thiamethoxam and cyromazine, when applied as seed treatments, were effective at controlling cabbage root fly eggs/larvae. However, neither treatment was as effective as the standard chlorpyrifos treatment (dose for dose).
- Carbosulfan granules applied to swede plots at drilling had little effect on cabbage root fly control over the period of the first fly generation.

## **Financial benefits**

- 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 £17M/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 about £25M per annum from the area of crop that needs protecting currently against attacks by the cabbage root fly.

### Action points for growers

#### Spinosad soil treatments (field experiment -swede)

• Seed treatment This naturally derived chemical can persist over two fly generations but the dose tested was insufficient to provide economic control of cabbage root fly larvae at any stage of the season.

#### *Insecticide treated baits (field experiment –swede and cauliflower)*

- Spinosad Did not reduce root damage or cabbage root fly numbers in either crop.
- **Chlorpyrifos** Similar effect to spinosad but some evidence of a reduction in stem damage on cauliflower.

#### Insecticide treated baits (glasshouse experiments - cauliflower)

- **Bait composition** Under glasshouse conditions, baits containing sugar only or yeast extract only were equally as effective at killing adult cabbage root flies as baits containing both. Baits did not need to be maintained in a liquid form (by addition of propylene glycol) to be effective.
- **Daylight** Fresh baits containing chlorpyrifos, dimethoate or spinosad were similarly effective when introduced to cages containing cabbage root flies but this efficacy declined rapidly after exposure to natural light for 5 days.

#### Novel insecticides (glasshouse experiment – cauliflower)

Module drench treatments

- Thiacloprid At 10 mg a.i./plant was almost as effective at controlling cabbage root fly larvae as the standard Dursban treatment (chlorpyrifos, 5 mg a.i./plant)
  Teflubenzuron Was as effective as the standard chlorpyrifos treatment at 0.4 mg a.i./plant
  Seed treatments
  Cyromazine Was as effective as the standard Gigant treatment (chlorpyrifos, 0.096 mg a.i./plant) between 0.5 and 1 mg a.i./plant. Some phytotoxicity observed at the highest dose tested (1 mg a.i./plant)
  Thiamethoxam Was as effective as the standard Gigant treatment between 0.5
- Thiamethoxam Was as effective as the standard Gigant treatment between 0.5 and 1 mg a.i./plant.

#### Natural insecticides (glasshouse experiment - swede)

• ECOguard granules Mortality of cabbage root fly eggs/larvae showed a positive dose response when eggs were placed around swede plants immediately after garlic granules had been applied. The effects diminished considerably when the granules were aged for 1 week and after 2 weeks the granules had virtually no effect.

Insecticide granules (field experiment – swede)

• **Carbosulfan** Failed to control first generation cabbage root fly larvae when applied to plots of swede at drilling.

## SCIENCE SECTION

## Introduction

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 involved field and glasshouse trials. Glasshouse trials can be used to estimate how much insecticide is required to give the desired level of control and results from glasshouse experiments pinpoint directly the treatments unlikely to be acceptable in commercial crops. Hence, a strong scientific base can be used to decide which new insecticides to include in a research programme.

Experiments were done to answer the following seven questions:

#### 1. Film-coated seed

How long do spinosad seed treatments persist?

#### 2. Insecticide treated baits

Will weekly spray applications of spinosad + bait control adult cabbage root flies in the field?

#### 3. Insecticide treated baits

Can the effectiveness of insecticide baits be improved and do female cabbage root flies that are exposed to insecticide baits lay eggs before they die?

#### 4. Insecticide treated baits

How does exposure to natural daylight for different periods affect the effectiveness of insecticide + bait mixtures?

#### 5. Film-coated seed and pre-planting module drench

How effective are the novel insecticides thiamethoxam, cyromazine, thiacloprid and teflubenzuron against cabbage root fly when applied as drench or seed treatments?

#### 6. 'Natural' insecticides

How long does the efficacy of Ecoguard garlic granules persist?

#### 7. Carbamate granules

Will carbosulfan granules applied at drilling control cabbage root fly larvae on swedes?

#### The seven 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: carbosulfan (Posse), cyromazine (Triguard), spinosad (Tracer), teflubenzuron (Nemolt), thiacloprid (Calypso), thiamethoxam (Actara) and garlic extract (ECOguard)

#### **Experiment 1.**

#### How long do spinosad seed treatments persist?

#### Materials and methods

#### a) Seed treatment

Swede seeds were film-coated with spinosad (Tracer 480SC) at a target loading of 100 g a.i./unit (1 unit = 100,000 seeds) and these seeds were used for the first drilling. Subsequent discussion with Dow Agrochemicals indicated that 72 g a.i./unit should be the maximum dose tested so a further batch of seed was treated at this dose and used for the other five drillings. A PVA sticker, at the rate of 2% of product weight, was applied with the treatment. The seeds were film-coated at HRI Wellesbourne and a further batch of seed was left insecticide-free.

#### b) Field experiment

Ten (1.83 m wide x 30 m long) seedbeds were prepared in the experimental area at HRI Wellesbourne. On 2 April, each seedbed was divided into two, 12 m long, plots to give a total of 20 plots. The 20 plots were divided into four replicate blocks of 5 plots. Each plot in a replicate block was assigned a drilling date at random. The first four plots were drilled on 4 April using a tractor mounted Stanhay Singulaire seed drill. Alternate rows were drilled with either insecticide-free or insecticide-treated seed at 13 seeds/m row and with 46 cm between rows. Further plots were drilled on 9 April, 24 April, 9 May, 23 May and 9 June respectively (Table 1).

The intention was to assess the efficacy of each treatment against a single generation of the cabbage root fly (either the first or second generation). Therefore, on each occasion, one half (6 m) of each plot was covered at drilling with Envirofleece® to exclude first generation cabbage root fly, whilst the other half of the plot was left exposed, but covered subsequently to exclude the second generation. On 7 May, 28 May and 7 July, seedling survival was assessed by counting the numbers of seedlings in a 3 m portion of each row. Then, on 7 July, approximately 6 weeks after the end of first generation (indicated by monitoring cabbage root fly egg laying in a nearby plot), the fleece was removed from the covered areas of drillings A-D to allow second generation flies access to the swedes. This fleece was then used to cover the areas that had been exposed to the first generation.

On 5 August, five plants were sampled from each of the insecticide-treated and insecticide-free rows in each plot that had been exposed to first generation cabbage root fly. A 15 cm diameter x 15 cm deep soil core was taken from around the roots of each plant. In addition, approximately 50 roots were harvested from each of the insecticide-treated and untreated rows. Cabbage root fly larvae and 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 mean root damage index was calculated based on the damage to each root, which was scored on a scale from 0 (no damage) to 4 (>50% damage).

On 21 August, all of the plots which had been exposed to second generation flies were covered with Envirofleece® to exclude the third generation. The fleece was removed from drilling F to allow egg laying by the third generation flies.

On 6 November, when all of the cabbage root fly larvae had formed pupae, the plots that had been exposed to the second generation were uncovered and samples were taken from these plots and from the drilling F plots and assessed as described above. The mean root damage index was calculated based on the damage to each root, which was scored on a scale from 0 (no damage) to 4 (>50% damage).

The seedling counts were square-root transformed before being were subjected to Analysis of Variance. Seedling survival between May and July was analysed using GLM analysis assuming a binomial distribution and a logit link function. The mean numbers of cabbage root flies recovered from the soil samples, the mean root weight and the mean root damage index were subjected to Analysis of Variance. The insect counts were square root transformed prior to analysis.

To provide background information, cabbage root fly egg laying was monitored in a small plot of cauliflower near to the main experimental plots. Soil samples were taken from 20 plants twice a week from 17 April until 7 October 2003. The eggs were extracted from the soil by flotation in water and counted.

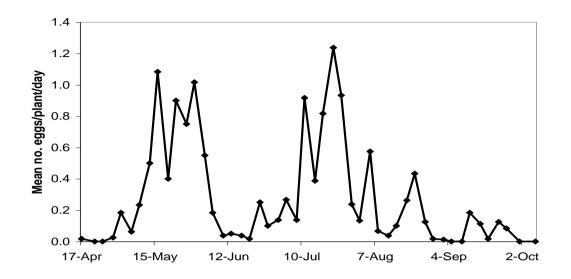
Table 1.Drilling dates for spinosad-treated swede seed indicating the cabbage root fly<br/>generations to which the treatments were exposed. Flies were excluded from<br/>the plots using fleece covers. Plots exposed to the first generation were<br/>uncovered between drilling and 25 June. Those exposed to the second<br/>generation were uncovered between 25 June and 21 August. Those exposed to<br/>the third generation were uncovered between 21 August and 5 November.

Treatment code	Drilling date	Fly generations assessed
A B	3 April 9 April	$1^{st}$ and $2^{nd}$ $1^{st}$ and $2^{nd}$
C C	24 April	$1^{st}$ and $2^{nd}$
D	9 May	$1^{\text{st}}$ and $2^{\text{nd}}$
E	23 May	$2^{nd}$
F	9 June	3 <sup>rd</sup>

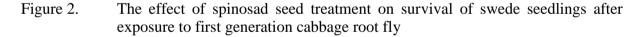
#### Results

Most first generation eggs were laid during the latter half of May (Figure 1). Egg laying by second generation cabbage root flies started to increase from late June onwards and peak numbers of eggs were laid in the third week of July. The third generation was less distinct than the other two generations, but appeared to start in mid August.

Figure 1. The numbers of cabbage root fly eggs laid per plant per day during 2003 in a monitoring plot of cauliflower at HRI Wellesbourne.



The seed treatment increased seedling survival during the first generation of cabbage root fly (p<0.001) (Figure 2). Root weight was increased (p=0.013) and the root damage index was reduced by a small but significant amount (p=0.033) following first generation fly attack (Figure 3). Second generation root damage could not be assessed due to the poor condition of the crop at harvest. The seed treatment consistently reduced the numbers of pupae recovered at all drilling dates against all three fly generations (p<0.001) (Figure 4). The best control (74% reduction in numbers of pupae) was achieved with the first drilling against the first generation of flies. However this treatment was applied at a rate of 100 g a.i./unit compared with the 72 g a.i./unit (66% reduction in numbers of pupae after 2<sup>nd</sup> drilling) used for the other five drillings. There appeared to be some reduction in seed treatment efficacy over time, but the pattern was not clear.



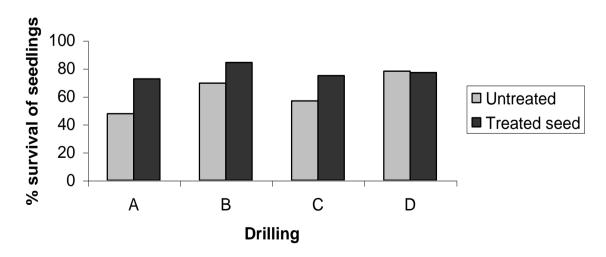
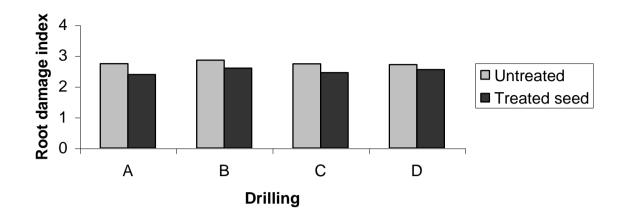
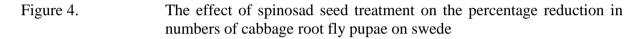
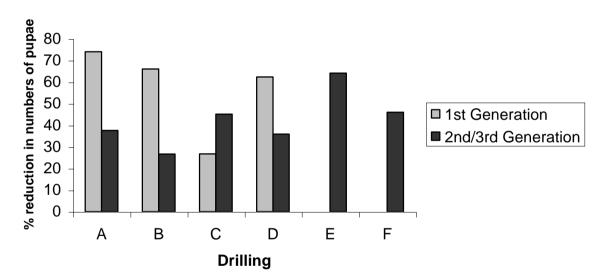


Figure 3. The effect of spinosad seed treatment on the root damage index of swede plants after exposure to first generation cabbage root fly







#### **Experiment 2.**

## Will weekly spray applications of spinosad + bait control adult cabbage root flies in the field?

#### Materials and methods

Three plots (7.5 m wide x 7.5 m long) were prepared in each of three fields (Cottage Field, Sheep Pens and Rush Pits) at HRI Wellesbourne. The three plots in each field were aligned on a north-south axis and were separated from each other by at least 20 m. On 9 July, 8 rows of glasshouse raised swedes and 7 rows of glasshouse raised cauliflowers were transplanted

into each of the 9 plots. The plants were spaced at 50 cm along the rows and 50 cm between rows. The plots were irrigated to establish the plants.

Immediately after planting, two batches (5 litre) of an insecticide bait solution were prepared consisting of 1000 g sucrose, 250 g yeast extract (Marmite), 500 ml Propylene glycol (to maintain the bait in liquid form) and 25 ml Codacide (to aid adhesion to the brassica leaves). Dursban 4 (5.2 ml) was added to one batch and Tracer (5.2 ml) was added to the other to give final concentrations of 500 mg/l of chlorpyrifos and spinosad respectively. One plot in each field was sprayed with the chlorpyrifos treated bait and one was sprayed with the spinosad treated bait. The third plot was left unsprayed. All sprays were applied using a knapsack sprayer fitted with a 1 m boom with 3 fine spray nozzles which delivered 200 l/ha at standard walking speed. At the spray rate used, the dose applied was equivalent to 100 g a.i./ha for both active ingredients. Using freshly prepared insecticide/bait solutions, the spray applications were repeated at weekly intervals, on 16 July, 23 July, 30 July, 6 August and 13 August respectively. The plots were left exposed to the local cabbage root fly population for a further week and then an area across the middle of each plot 3 plants (swedes and cauliflowers) wide x 7.5 m long, was covered with fleece to prevent further invasion.

On 6 November, when nearly all of the cabbage root fly larvae had finished feeding and formed pupae, the plots were uncovered and 15 cm diameter x 15 cm deep soil cores were taken from around the roots of 10 plants of each crop in each plot. The cabbage root fly larvae and pupae were extracted from the soil samples by flotation in water and the numbers of larvae and pupae recovered were recorded. The plant roots were washed, weighed and scored for cabbage root fly damage. The roots were scored on a 0 - 3 scale where 0 = no damage, 1 = superficial damage, 2 = moderate damage and 3 = unmarketable. Most of the damage to cauliflowers appeared to be in the lower part of the plant stem, so the number of cabbage root fly mines in this area was also assessed.

The mean numbers of cabbage root flies recovered from the soil samples, the mean root weight, the mean root damage index and the mean numbers of mines in cauliflower stems were subjected to Analysis of Variance. The insect counts were square root transformed prior to analysis.

#### **Results**

The mean numbers of pupae recovered (all treatments) were similar in the 3 fields, ranging from 10 pupae/plant in Rush Pits to 14.5 pupae/plant in Sheep Pens. There were no consistent differences between treatments in the numbers of pupae recovered from either crop (p=0.499) (Figure 5). Similarly, the mean root damage score was not reduced by either insecticide treatment when compared with the insecticide-free control (p=0.406 – cauliflower; p= 0.381 – swede) and there was no effect on root weight (p=0.07 – cauliflower; p=0.46 - swede). However, there was some evidence of a reduction in the numbers of cabbage root fly mines in the stems of cauliflower plants treated with chlorpyrifos when compared with the other two treatments (p=0.006) (Figure 6).

Figure 5. The mean numbers of cabbage root fly pupae recovered from around swede and cauliflower plants sprayed at weekly intervals with insecticide treated baits.

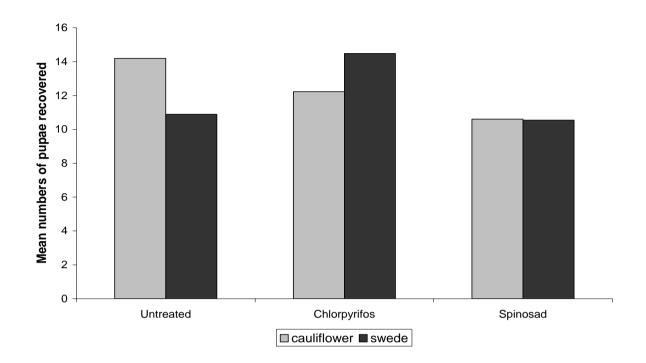
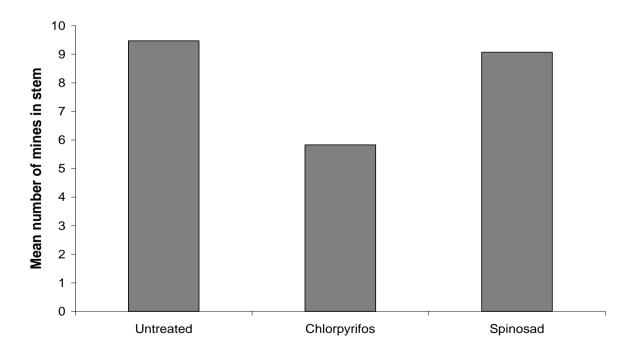


Figure 6. The mean numbers of cabbage root fly mines in cauliflower stems after plants were sprayed at weekly intervals with insecticide treated baits.



#### **Experiment 3.**

## Can the effectiveness of insecticide baits be improved and do female cabbage root flies that are exposed to insecticide baits lay eggs before they die?

#### Materials and methods

Cauliflower plants (cv Lateman) were grown in Optipot 9M pots containing Levington compost and were maintained under glasshouse conditions. These plants were used for all the experiments.

One insecticide, spinosad (Tracer), was tested at a single dose (320 mg a.i./l). This dose was lower than the doses used in Experiments 2 and 4 (500 mg a.i./l) and was chosen because it was more likely to facilitate discrimination between the efficacy of the different bait mixtures than the higher dose. A range of bait solutions (Table 2) was prepared to assess the effects of sugar and protein concentrations on the effectiveness of the baits. The effects of removing propylene glycol (used in previous trials to maintain the bait in liquid form) were also assessed. From 25 February to 8 March 2004, a series of trials was done in a glasshouse. All treatments were sprayed onto cauliflower plants and left overnight to allow the residues to dry onto the leaves. Each trial consisted of one plant treated with each insecticide-treated bait and an untreated control plant sprayed with insecticide-free bait. Fresh bait was prepared for each trial and each insecticide-treated bait was tested on three occasions.

Table 2	Composition of bait solutions assessed against the adult cabbage root flies
	under glasshouse conditions

Treatment	Sucrose	Yeast extract	Propylene glycol
	(% w/v)	(%w/v)	(v/v)
1 (untreated)	20	5	10
2	20	0	10
3	0	5	10
4	20	5	10
5	5	5	10
6	20	1	10
7	20	5	0
8	10	2	10
9	10 (honey)	0	10
10	10 (honey)	0	0

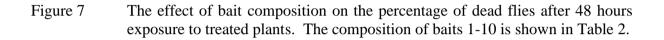
Fifteen male and fifteen female cabbage root flies (4-8 days old) were placed into nylon net cages (35 cm x 35 cm x 35 cm) and transferred to an illuminated glasshouse (set to give a 16-hour day length) on the day the cauliflower plants were treated. A dish containing sucrose solution absorbed onto cotton wool, together with a bottle containing fresh water and a blotting paper wick, were placed into each cage. The following morning, a layer of dry, sieved soil was placed over the surface of each cauliflower pot. The numbers of dead flies of each sex were recorded before one treated plant was placed into each cage. The numbers of dead flies of each sex were then recorded after 6, 24 and 48 hours. After the final assessment of mortality, the soil was washed from the top of each pot into a plastic container. Water was added and the soil was

agitated to allow flotation of the cabbage root fly eggs. The numbers of eggs laid in each of the pots were recorded.

The egg counts were square root transformed and subjected to Analysis of Variance, whilst fly mortality was analysed using GLM, assuming a binomial distribution and logit link function.

#### Results

All of the bait solutions killed more adult cabbage root flies than the untreated control (bait 1) (p<0.001). After 48 hours more than 76% of male and 91% of female flies had been killed by the treated baits (Figure 7). However, there was no difference between treated baits. Egg laying was unaffected by any of the treated baits compared with the untreated control (p=0.895) (Figure 8).



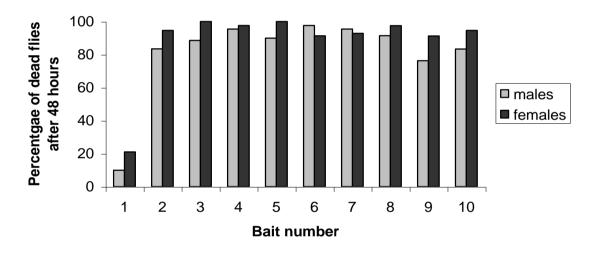
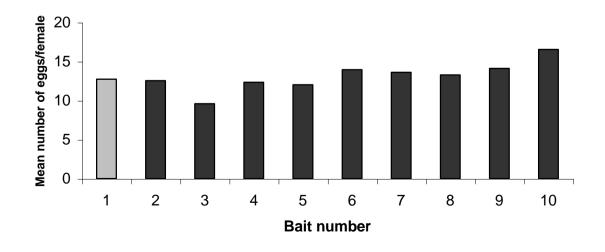


Figure 8 The effect of bait composition on the mean number of eggs laid/female after exposure to treated plants. The composition of baits 1-10 is shown in Table 2.



#### **Experiment 4.**

## How does exposure to natural daylight for different periods affect the effectiveness of insecticide + bait mixtures?

#### Materials and methods

Cauliflower plants (cv Lateman) were grown in Optipot 9M pots containing Levington compost and were maintained under glasshouse conditions. These plants were used for all the experiments.

Three insecticides (spinosad (Tracer), chlorpyrifos (Dursban) and dimethoate (Dimethoate 40)) were tested at a single dose (500 mg a.i./l). The bait solution used was the same as that used in the field experiment (Experiment 2) and contained sucrose (20% w/v), yeast extract (5% w/v), propylene glycol (10% v/v) and codacide (0.5% v/v).

On 4 September 2003, 15 plants were sprayed with freshly prepared bait solution containing either spinosad, dimethoate, chlorpyrifos or no insecticide. A layer of dry, sieved soil was placed over the surface of each pot and three plants from each treatment were placed immediately into nylon net cages (35 cm x 35 cm x 35 cm) and transferred to an illuminated glasshouse (set to give a 16-hour day length). A dish containing sucrose solution absorbed onto cotton wool together with a bottle containing fresh water and a blotting paper wick were placed into each cage. Fifteen male and fifteen female cabbage root flies (4-8 days old) were placed into each cage soon after the plants had been introduced and then the numbers of dead flies of each sex were recorded after 24 and 48 hours. Following the final assessment of fly mortality, the soil from the top of each pot was rinsed into a plastic container. The soil was agitated to allow flotation of the cabbage root fly eggs and the numbers of eggs laid in each pot were recorded.

To provide test plants with aged residues, the remaining plants were placed in a cold frame and covered with insect-proof netting to exclude wild cabbage root flies. The glass cover to the frame was left open during daylight hours, but closed at night or when rain was forecast. The experiment was repeated after the residues had aged for 5 and 11 days.

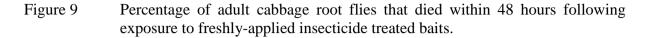
On 15 September, the plants were treated with fresh applications of insecticide + bait solution. Once again, the residues were allowed to age and the plants were tested 7 and 14 days after the second treatment.

The mean numbers of cabbage root fly eggs recovered from the pots and the percentage of dead flies were subjected to Analysis of Variance. The egg counts were square root transformed and the percentage mortalities arcsine transformed prior to analysis.

#### <u>Results</u>

Fresh residues of all three test insecticides killed both male and female cabbage root flies rapidly (p<0.001; Figure 9). After 48 hours there was 100% mortality. The rate of kill was such that egg laying was almost eliminated in cages containing treated plants (p=0.004; Figure 11). After the residues had aged for 5 days under natural light, the rate of kill diminished (Figure 10) and consequently egg laying was reduced by only about 25% in all treatments compared with

untreated plants. After the second application had been aged for 7 days there was little or no effect from any of the treatments.



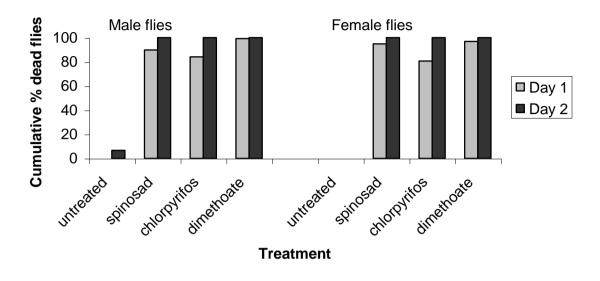
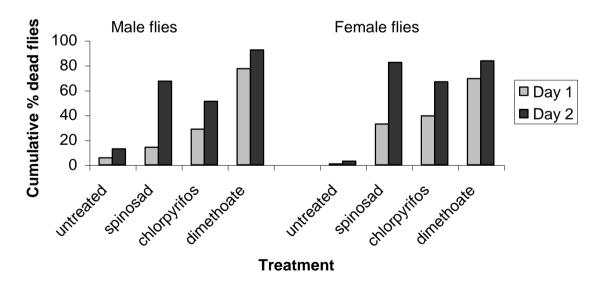
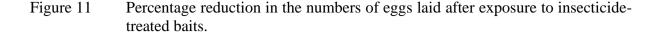
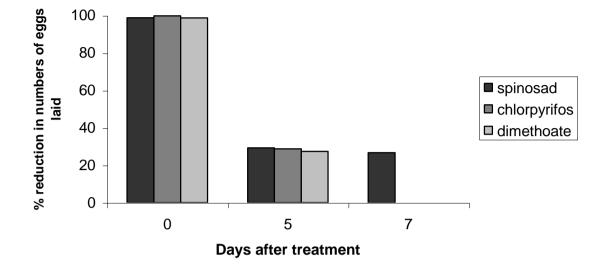


Figure 10 Percentage of adult cabbage root flies that died within 48 hours following exposure to insecticide treated baits which had been aged for 5 days.







#### **Experiment 5.**

How effective are the novel insecticides thiamethoxam, cyromazine, thiacloprid and teflubenzuron against cabbage root fly when applied as drench or seed treatments?

a) Drench treatment

#### Materials and methods

Two insecticides (thiacloprid (Calypso) and teflubenzuron (Nemolt)) were evaluated as drench treatments. On 9 December 2003, untreated swede seed was sown in 308 Hassy trays containing Levington compost. On 16 January 2004, 5 sets of 14 plants (1 row of modules) were transferred to a clean Hassy tray, leaving two empty rows between each set of plants. Each set of plants was treated with one of five doses of one of the test insecticides. The process was repeated for each of the two test insecticides. Each treatment was applied by adding 1 ml of a solution in water using a laboratory pipette. The doses applied were 0.4, 1, 2, 5 and 10 mg a.i./plant for both insecticides. Fourteen plants were also treated with chlorpyrifos at the rate recommended currently (5 mg a.i./plant), as a positive control. In each case, the insecticide solution was washed into the peat with a similar volume of clean water, immediately after the insecticide had been applied. One Hassy tray was left untreated.

On 19 January 2004, 3 days after application of the insecticide treatments, 12 plants from each treatment were transplanted into Optipot 9M pots filled with John Innes No. 2 compost. The newly-potted plants were moved into a glasshouse compartment maintained at  $20 \pm 2$  °C. The treatments were arranged in 12 blocks, each containing 1 plant from each of the 12 treatments. The positions of the plants within each block were randomised fully. The plants were allowed to establish for 2 weeks.

On 2 February, twenty cabbage root fly eggs, obtained from the laboratory culture maintained at HRI Wellesbourne (Finch & Coaker, 1969), were washed onto the soil at the base of each plant. After one month at 20 °C, most of the cabbage root fly should have formed pupae, so from 2 March, batches of the pots were taken from the glasshouse so that the fly pupae could be extracted. The cabbage root fly pupae were extracted from the potting compost by flotation in water. Records were taken of the numbers of pupae found and of root weight.

#### b) Seed-treatment

#### Materials and methods

Cauliflower seeds were film-coated at HRI Wellesbourne. Seeds were treated with chlorpyrifos (Gigant), cyromazine (Triguard 75WP) and thiamethoxam (Actara 25WG). As chlorpyrifos is liquid at room temperature, the chlorpyrifos (positive control) formulation was applied, at the commercially recommended rate of 9.6 g a.i./unit (1 unit = 100,000 seeds), by using a slurry with Talcum powder. Each of the test insecticides was applied at target loadings of between 1 and 100 g a.i./unit using a PVA sticker at a rate of 2% of product weight. A further batch of seed was left insecticide-free.

On 9 December 2003, the treated swede seed was sown (42 seeds/treatment) in 308 Hassy trays containing Levington compost. A tray of untreated seed was also sown. The trays were maintained under glasshouse conditions ( $20 \pm 2$  °C). On 19 January 2004, 12 plants from each treatment were transplanted into Optipot 9M pots filled with John Innes No. 2 compost. The treatments were arranged in 12 blocks each containing one plant from each of the 12 treatments. The positions of the plants within each block were randomised fully. The plants were allowed to establish for two weeks.

On 2 February, twenty cabbage root fly eggs, obtained from the laboratory culture (Finch & Coaker, 1969), were washed onto the soil at the base of each plant. After one month at 20 °C, most of the cabbage root fly should have formed pupae, so from 2 March, batches of the pots were taken from the glasshouse so that the fly pupae could be extracted. The cabbage root fly pupae were extracted from the potting compost by flotation in water. Records were taken of the numbers of pupae recovered and of root weight. The numbers of pupae recovered were subjected to Analysis of Variance. The data were arcsine transformed prior to analysis.

#### Results

Almost 70% of the eggs that were inoculated were recovered as larve/pupae from the insecticide-free control plants. The chlorpyrifos drench treatment (Dursban) reduced larval/pupal recovery by 99.6% and the chlorpyrifos seed treatment (Gigant) by 85.2%. With the exception of teflubenzuron, there was a clear dose response to the experimental insecticide treatments (p<0.001) (Figures 12 & 13).

At 10 mg a.i./plant thiacloprid was almost as effective at controlling cabbage root fly larvae as the standard Dursban treatment (chlorpyrifos, 5 mg a.i./plant). Teflubenzuron was as effective as standard chlorpyrifos treatment at 0.4 mg a.i./plant. As a seed treatment, cyromazine was as effective as standard Gigant treatment (chlorpyrifos, 0.096 mg a.i./plant) between 0.5 and 1 mg a.i./plant. Some phytotoxicity was observed at the highest dose tested (1 mg a.i./plant). Thiamethoxam was as effective as the standard Gigant treatment between 0.5 and 1 mg a.i./plant. Thus neither treatment was as effective as the standard chlorpyrifos treatment (dose for dose).

Figure 12 Percentage reduction in the numbers of cabbage root fly larve/pupae recovered from plants treated with insecticide drench treatments compared with insecticide-free control plants. Plants were treated with chlorpyrifos (Dursban), at the rate recommended currently (5 mg a.i./plant), as a positive control.

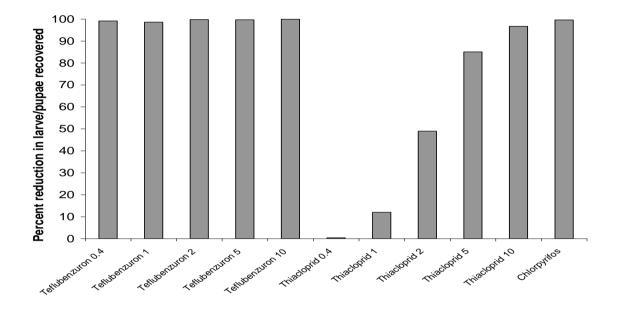
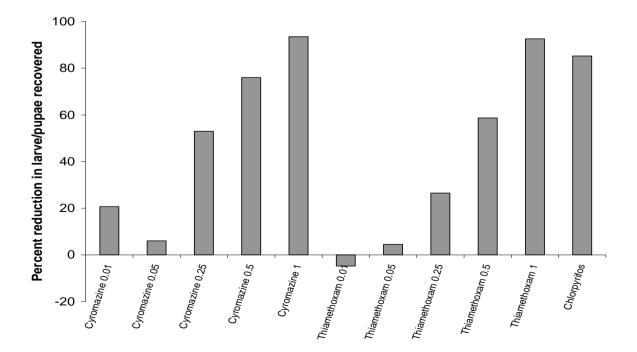


Figure 13 Percentage reduction in the numbers of cabbage root fly larve/pupae recovered from plants treated with insecticide seed treatments compared with insecticide-free control plants. Seeds treated with chlorpyrifos at a rate of 9.6 g a.i./unit (1 unit = 100,000 seeds) were used as a positive control.



#### **Experiment 6.**

#### How long does the efficacy of Ecoguard garlic granules persist?

#### Materials and methods

Three doses of garlic granules were selected, based on results obtained in 2002 (Jukes *et al*, 2003). These doses had killed some larvae, but mortality rates were less than 100%. The doses were 0.12, 0.25 and 0.5 g product/plant.

Swede seeds were sown into 308 Hassy trays and grown in a heated glasshouse under lights. At the 4-leaf stage, 160 plants were transplanted into Optipot 9M square pots and allowed to establish under the same glasshouse conditions  $(20 \pm 2 \,^{\circ}C)$  for a further 4 weeks.

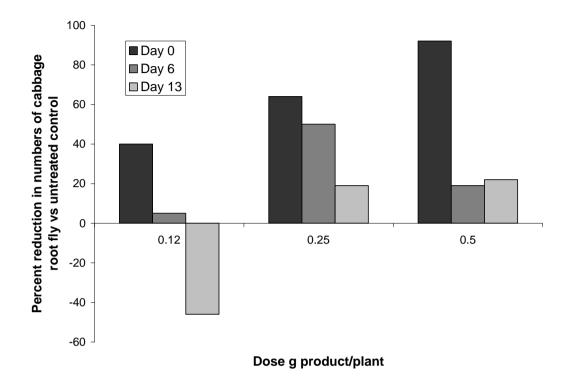
On 16 December 2003, pre-weighed samples of garlic granules were applied to the base of each of the swede plants (40 plants treated at each dose) and 40 plants were left untreated. The granules were covered with loam soil, which was moistened with water (1-2 ml). Immediately after treatment, twenty cabbage root fly eggs, obtained from the laboratory culture maintained at HRI (Finch & Coaker, 1969), were washed onto the soil at the base of 8 plants from each treatment. The process was repeated on other sets of 8 plants 6, 13, 20 and 27 days after treatment. The experimental plants were arranged in 8 blocks, each containing 1 plant from each of the 20 treatments (4 doses x 5 times). The positions of the plants within each block were randomised fully.

On 19 January 2004, the cabbage root fly larvae and pupae from the first inoculation were extracted from the potting compost by flotation in water and the numbers recovered from each plant were recorded. The roots were washed, weighed and scored for cabbage root fly damage. Subsequent inoculations were washed out at weekly intervals thereafter. The mean numbers of cabbage root flies recovered from the soil samples were square root transformed and subjected to Analysis of Variance.

#### <u>Results</u>

Garlic dose had a statistically significant effect (p=0.012) on the numbers of cabbage root fly that survived to pupation. When cabbage root fly eggs were inoculated on the same day that the garlic granules were applied, doses of 0.12, 0.25, 0.5 product/plant reduced cabbage root fly numbers by 40, 64 and 92% respectively compared with the untreated control (Figure 14). Efficacy of the granules declined as they aged (p<0.001) and even within one week, control of cabbage root fly was reduced considerably. There was also a statistically significant interaction between dose and inoculation date (p<0.001).

Figure 14. Percentage reduction in the numbers of cabbage root fly pupae after treatment with garlic granules compared with survival on the untreated control plants.



#### **Experiment 7.**

#### Will carbosulfan granules applied at drilling control cabbage root fly larvae on swedes?

#### Materials and methods

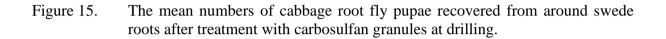
Two (1.83 m wide x 50 m long) seedbeds were prepared, at either side of Experiment 1, in the experimental area at HRI Wellesbourne. On 9 May 2003, the beds were drilled with swede seed, at 13 seeds/m row, using a tractor mounted Stanhay Singulaire seed drill. Four rows were drilled into each bed to give a row spacing of 46 cm. Carbosulfan granules (Posse 10G, 70 g product/100m row) were added to all four rows in each bed at drilling using a Horstine Farmery granule applicator. The granules were placed onto the soil surface and incorporated to a depth of about 5 cm by the following seed drill. The dose rate chosen is the recommended rate for application of Posse 10G to brassicas. The plots were left uncovered, so that they were exposed to egg laying by the first generation of cabbage root fly.

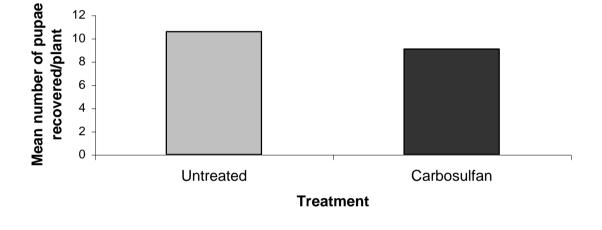
On 16 July 2003, 15 cm diameter x 15 cm deep soil cores were taken from around the roots of 10 plants in the central 2 rows of each treated bed. In addition, approximately 50 additional roots were harvested from each bed. Cabbage root fly pupae were extracted from the soil samples by flotation in water and the numbers of insects recovered were recorded. The roots were washed, weighed and scored for cabbage root fly damage. The root damage index was calculated based on scoring damage to individual roots on a scale from 0 (no damage) to 4 (>50% damage). The mean numbers of cabbage root flies recovered from the soil samples were square root transformed and subjected to Analysis of Variance. The results were compared with

samples from the adjacent insecticide-free plots in Experiment 1. The mean root damage indices from each treatment were compared using Chi squared analysis.

#### Results

Root damage due to feeding by cabbage root fly larvae was heavy on all plots. Compared with the insecticide-free control, the carbosulfan treatment reduced the numbers of insects by a small but statistically insignificant amount (p=0.623) (Figure 15). Chi squared analysis showed that there was no reduction in root damage due to the carbosulfan treatment (P>0.05). However, visual inspection of the plots during the summer indicated that the carbosulfan treatment reduced aphid and flea beetle damage to the foliage.





## CONCLUSIONS

#### Spinosad - film-coated seed

In 2002, spinosad seed treatment at 150 g a.i./unit provided excellent control of first generation cabbage root fly larvae and was still partially effective against second generation larvae (12-16 weeks after drilling) (Jukes *et al.*, 2003). In 2003, lower doses of spinoad were applied (100 g ai./unit for the first drilling and 72 g a.i. for subsequent drillings). Even at these lower doses there was some cabbage root fly control at all drillings and the higher dose used on the first drilling produced a 74% reduction in numbers of first generation larvae. Control of first generation larvae was generally better than second generation larvae but there was no clear pattern of decreasing efficacy over time. The results confirm that spinosad seed treatment shows potential for being an effective first generation control measure, both in terms of reduction in root damage and increase in seedling stand. However, the dose may need to be higher both for effective first generation control and partial control of the second generation. By inference, the results suggest that the seed treatment alone would provide adequate protection to module raised leafy brassicas, where lower levels of control are required.

#### Insecticide treated baits

Glasshouse trials with insecticide-treated baits (Jukes *et al*, 2001) showed that it is possible to increase the amount of insecticide acquired by adult onion flies and cabbage root flies using this method of application, thereby killing them. The questions asked in 2003-4 were:

- 1. Can the effectiveness of insecticide baits be improved and do female cabbage root flies that are exposed to insecticide baits lay eggs before they die? (Experiment 3)
- 2. How does exposure to natural daylight for different periods affect the effectiveness of insecticide + bait mixtures? (Experiment 4)

In Experiment 3, all of the treated baits tested performed similarly and female and male flies died at similar rates. Bait excluding sugar and bait excluding protein were as effective as bait containing both sugar and protein. The type of sugar had no effect. Bait excluding propylene glycol (added to keep bait in liquid form) was as effective as bait with propylene glycol and there was little difference in egg laying between untreated and treated plants.

Experiment 4 showed that at a higher dose (500 mg a.i./litre), fresh insecticide + bait residues killed female flies so rapidly that they were unable to lay eggs. However, as the residues aged, fly mortality decreased and as a consequence, egg-laying increased.

Results from the plot trials to evaluate mid-season sprays of spinosad or chlorpyrifos + bait were disappointing. Little or no reduction in insect survival was observed, even though weekly sprays were applied. However, there was some evidence that chlorpyrifos reduced damage to the lower part of cauliflower stems. The failure of the bait treatments in the field is explained by the results of Experiment 4, which showed that although fresh residues killed quickly and very few eggs were laid, aged residues lost potency quickly and the rate of kill decreased. After 5 days, egg laying on treated plants was reduced by only about 25% compared with untreated plants.

From the results of the field and laboratory experiments done during 2003-4 it would appear that the insecticide + bait treatments are not sufficiently persistent to provide effective control in the field and that improvements would only be likely if a formulation were developed to increase the stability of such treatments considerably.

#### Novel insecticides

The four 'new' insecticides were evaluated against cabbage root fly as either drench (thiacloprid, teflubenzuron) or seed (thiamethoxam, cyromazine) treatments. Chlorpyrifos applied either as a drench (Dursban) or seed treatment (Gigant) was used as the postivie control treatment. All the insecticides controlled cabbage root fly and with the exception of teflubenzuron, there was a clear dose response to the experimental insecticide treatments. The behaviour of thiacloprid was similar to the closely related compound imidacloprid and the behaviour of teflubenzuron was similar to diflubenzuron (Jukes *et al.*, 2001). These results confirm that apart from spinosad, several other active ingredients appear to work effectively as soil insecticides against cabbage root fly and have the potential to be used as drench or seed treatments on leafy brassicas. The key question is 'how can such soil-active insecticides be applied so that they are in the right location to protect long-season crops such as swede'?

#### Garlic granules

Experiments done in 2002-3 showed that targeted soil application of the granular product was effective against cabbage root fly eggs and larvae under glasshouse conditions (Jukes *et al.*, 2003). Although the efficacy of fresh garlic granules was confirmed in the current project (Experiment 6), as the granules aged, their efficacy declined and even after 6 days, control of cabbage root fly larvae was reduced considerably.

#### Carbosulfan granules

Carbosulfan (Posse), like benfuracarb (Oncol 10G) (Jukes *et al.*, 2003) does not have the same efficacy against cabbage root fly larvae as carbofuran (Yaltox). In the single dose trial conducted in this project (70 g product/100 m) there was some evidence of a reduction in the numbers of cabbage root fly larvae present, but this was insufficient to reduce levels of root damage. However, carbosulfan could be an effective treatment at drilling for the control of flea beetles and aphids.

## **TECHNOLOGY TRANSFER**

Results from the project were presented and discussed at the following events:

11-Dec 2003	HDC swede meeting – HRI Wellesbourne
12-Dec 2003	PSD meeting on chlorpyrifos
14-Jan 2004	Brassica Growers Association Conference
03-Feb 2004	HDC Roadshow Scotland
17-Feb 2004	HDC Roadshow Kent
10-Mar 2004	HDC Roadshow Lancashire
17-Mar 2004	HDC Roadshow Cornwall
21-Apr 2004	BGA technical meeting
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Articles:

Collier, R.H. (2003). Cabbage root fly control: the future. HDC News 93, June 2003, 21-23.

Collier, R.H. (2003). No quick fix. Grower, December 11 2003, 22-23.

Jukes, A.A. & Collier, R.H. (2004). Does cabbage root fly rise to the bait? HDC News March 2004, 12-13.

### 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**

We thank the HDC for funding this work and the two project co-ordinators, Mr Ian Morrison of Kettle Produce Ltd, Fife and Mr Fred Tyler of Alphagrow Ltd, Lancashire, for the information they supplied and for the considerable enthusiasm and encouragement they provided throughout the project. We are also grateful to the manufacturers who supplied us with samples of their products. Statistical analyses were done by Andrew Mead, HRI.

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