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AUTHENTICATION

We declare that this work was done under our supervision according to the procedures described herein and that the report represents a true and accurate record of the results obtained.

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GROWER SUMMARY

Headline

Chard and spinach leafminer damage can be reduced to commercially acceptable levels by using a combination of monitoring, cultural practices and insecticides.

Background

Growers of high value leafy salads use mesh netting to protect some of their crops from a complex of insect and vertebrate pests and this has proved commercially effective against the leafminer species that can damage leafy salad Brassicas (HDC project FV 301). It is impractical, however, to use netting to protect chard and spinach, which can cover much larger areas, but, more importantly, can also be prone to damage by the netting. These particular crops were vulnerable to the recent increase in pest pressure from leafminer species therefore effective methods of managing the problem were required urgently.

This project aimed to investigate the possibility of improved insecticide-based control methods for chard and spinach leafminers and to disseminate the new knowledge to growers. The specific objectives were to:

- i) rear leafminer species collected from chard and spinach crops in the SE and NE of England;
- ii) develop a monitoring methodology for chard and spinach leafminers;
- iii) evaluate seven insecticide-based management treatments in a field trial;
- iv) disseminate new information to growers.

Summary

In 2011, the only leafminer pest species of any significance that was found to be attacking chard and spinach crops was the beet leafminer or mangold fly” *Pegomya hyoscyami*.

Various sampling methods were used to collect leafminers and the most effective was collecting pupae from the soil beneath mined plants early in the year. Removal of volunteer chard and spinach plants, particularly early in the year would therefore be an effective cultural control method for helping to reducing pest pressure.

Mass-rearing and monitoring data showed that there are probably only three generations of *P. hyoscyami* in most years. This means that significant egg-laying by the mangold fly is

therefore most likely to occur in south England during late May to mid-June and from mid-August to September.

A comparison of monitoring methods found that the most effective and direct technique would be to plant a small area of 'trap' red chard at monthly intervals, positioned near the main crops) and check these regularly for the presence of the distinctive white eggs.

Of the actives tested, Decis Protech (deltamethrin), the coded product HDCI 015 and Gazelle (acetamaprid) all reduced leafminer damage substantially and prevented larvae maturing to the later instars when they become large white maggots.

No evidence of resistance to insecticides was detected. The effective actives all have different modes of action, which is very encouraging, because this should reduce the risk of resistance development and thus ensure the longer-term sustainable production of spinach and chard crops, even in *P. hyoscyami* 'hot-spots'.

Financial Benefits

The project has delivered significant financial benefits, because it proposes a low-risk management method for the mangold fly that does not rely on the use of nets. The research has also shown that several insecticides, both currently available and a coded product near registration, will prevent the development of large leafminer maggots in the crop. The full financial consequences of having crops rejected, or lost, due to the presence of leafminer maggots or mines is difficult to quantify precisely, but the estimated sales value of spinach and chard ranges between £10,000 to £20,000 per hectare. The funding provided by the HDC was £22,788.00 and so within a single season, the full value of the project could be recovered by avoiding the damage caused by leafminer and maintaining the sales value of only 1 to 2 hectares of crop.

Action Points

- Removal of volunteer chard and spinach plants in spring should be encouraged to reduce host availability for leafminer populations emerging from winter diapause.
- Early preparation of fields to plough-in mined leafminer host plants, should reduce populations by killing pupae present in the soil.

- To prevent insecticide-resistance development and thus ensure sustainable long-term chard and spinach production, a leafminer monitoring system should be put into practice. This will enable a reduction in the number of sprays applied to crops.
- When the monitoring data indicates the need for sprays, the use of available actives should be alternated. They have different modes of action and so this will also help prevent resistance development.

SCIENCE SECTION

Introduction

Prior to the wide-spread adoption of insecticide-treated seed, leafminers of Chenopodiaceae crops were serious and chronic pests. Widespread use of seed treatments, such as Nuprid 600FS for sugar beet (*Beta vulgaris* subsp. *vulgaris*), may explain the reduced leafminer damage in recent years, but their apparent success has meant that there has been an associated lack of research on this important UK pest complex.

The leafminers that cause economic damage to Swiss chard (*Beta vulgaris* subsp. *cicla*) and spinach (*Spinacea oleracea*) (Chenopodiaceae), differ from the species that attack leafy salad brassicas. Spinach and chard in the UK are damaged by at least five Diptera (fly) species: *Pegomya hyoscyami* (the beet leafminer or “mangold fly”), *Amauromyza flavifrons*, *Delia echinata* (the spinach stem fly), *Pegomya betae* and *Clanoneurum cimiciforme*. These leafminers have relatively wide host-plant ranges and so, in addition to attacking commercial crops, populations can develop on native weed species such as *Chenopodium album* (fat hen).

The type of economic damage caused by leafminers falls into two categories. Mines caused by their larvae are unsightly and reduce the value of the crop. A worse scenario occurs, however, when the mines go unnoticed and the larvae are able to feed for about two weeks. These late instar larvae are henceforth referred to as maggots, whose presence in speciality salad crops is completely unacceptable and so leaves the crop unmarketable (Figure 1).



Figure 1. A fully developed *Pegomya hyoscyami* (the beet leafminer or mangold fly), maggot (also known as a late instar larva) with mine damage to the leaf on the right hand side of the image.

The damage caused by the different leaf miners can, to a certain extent, be used to identify the species responsible. *A. flavifrons* causes large blotch shaped mines. *D. echinata*, excavates spinach stems as well as mining the leaves, while the blotch mines of *P. betae* generally occupy an entire leaf. *P. hyoscyami* creates a large blotch mine, often containing several larvae (Figure 2).



Figure 2. Many *P. hyoscyami* larvae occupying a large mine made in a red chard leaf.

These larvae turn into pupae, which remain in the soil for 2-3 weeks during the summer, before emerging as adult flies. At the start of the project, the literature suggested that they could have more than three generations per year and so populations could reach high numbers in late season, particularly if the summer weather was hot and dry.

As there were probably several species involved and eggs are laid in the crop by influxes of immigrant adult flies, one of the main difficulties with managing these species is the sporadic nature of the problem. In addition, once the larvae have created mines, they then occupy a protected environment, where they then might only be reached by systemic insecticides or parasitoids. The possible insecticides available for use on these crops, therefore, need to be used in a selective and informed manner, which could be facilitated by

research aimed at developing improved monitoring techniques.

Growers of high value leafy salads use mesh netting to protect some of their crops from a complex of insect and vertebrate pests and this has proved commercially effective against the leafminer species that can damage leafy salad brassicas (FV 301). It is impractical, however, to use netting to protect chard and spinach, which can cover much larger areas and which can also be prone to damage by the netting. These particular crops, therefore, remain vulnerable to the recent increase in pest pressure from leafminer species and new methods of managing this problem are required urgently.

This one-year-duration project aimed to investigate the possibility of improved insecticide-based control methods for chard and spinach leafminers and to disseminate the new knowledge to growers. The specific objectives were to, i) rear leafminer species collected from chard and spinach crops in the SE and NE of England; ii) develop a monitoring methodology for chard and spinach leafminers; iii) evaluate seven insecticide-based management treatments in a field trial; iv) disseminate the new information to growers.

Materials and methods

Collection of leafminer species

Several methods were adopted to ensure that the project established colonies of the different leafminer species possibly responsible for the threat to spinach and chard crops. (i) Adult flies were collected using a sweep-net from chard crops at Intercrop Ltd. and released onto caged chard plants. (ii) Crops and weed species were scouted to collect leaf mines and the distinctive elongate, white eggs of *P. hyoscyami* (Figure 3).



Figure 3. The characteristic, easily recognisable, batches of 1-6 eggs laid by female *P.*

hyoscyami leafminers (LHS image). A close-up view of *P. hyoscyami* eggs (RHS image).

In addition, at the start of the summer in April 2011, the soil underneath mined volunteer chard and *C. album* (fat hen) plants was excavated to search for pupae. (iii) Small 'trap' plants and plots of chard were planted at bi-monthly intervals over the summer and taken to Intercrop Ltd and an organic allotment in Medway. These were monitored regularly and collections of leafminer eggs made from them.

It was assumed that most of the leafminer adults would be immigrants from the vegetation surrounding the Intercrop Ltd. Blue (attractive colour to Diptera) sticky traps, therefore, were placed at the edges of chard and spinach crops from April and May 2012 to sample the adult-fly population.

A visit was also made to the Emmett (Nottinghamshire) farm (NG22 8TW) in late summer (20th September 2011) to assess leafminer pressure and collect samples.

Mass rearing of leafminers

The leafminer rearing work took place using the entomological facilities at the Natural Resources Institute, Chatham Maritime. This work had three aims, which were to obtain, (i) biological information on the life cycles and the number of generations that occur annually for these species, (ii) macro-photographic images of the adults and the type of damage (characteristic mines) caused by the larvae to chard and spinach leaves, and (iii) generate the large number of adult insects required for the field trial that was planned to involve plots on which leafminer adults were caged.

Prior to making the leafminer collections, approximately 400 Swiss red-chard plants were sown to provide the diet for the developing larvae. These plants were placed into insect cages (1 m x 35 cm x 50 cm) (Figure 4), to provide a contained environment for rearing the leafminers. Adult flies captured by sweep-net were then released into cages with a source of honey solution for adult feeding. Leaves with mines or eggs collected from the field were placed onto the surfaces of caged plants' leaves and left for the insects to emerge and to move onto the healthily growing leaves of the experimental plants. Leafminer pupae collected from the soil at Intercrop Ltd were kept in an insect cage containing a healthy Swiss chard plant and a source of honey solution. The emerging adult flies were allowed to feed and oviposit (lay eggs) on the chard plants.



Figure 4. The type of cage used to allow sampled flies to oviposit on red chard plants. This cage type was also used to obtain oviposition on the experimental bioassay plants.

To avoid the leafminer larvae killing the colony plants, the number of eggs or larvae per plant was limited to between 15 and 20. To achieve this, plants were replaced regularly in the insect cages and the plants covered with perforated, plastic “bread bags”. This containment was necessary, because the late instar leafminer larvae were mobile, especially at night, and emerged from their mines to crawl between plants and leaves. In addition, at the end of the larval stage, most larvae emerged from their mines and migrated down to the soil to pupate.

Macro-photographic images of the adults and the type of damage (characteristic mines) caused by the larvae to chard and spinach leaves were taken using a Canon MP-E 65 mm (f/2.8 1-5x) macro lens.

Field trial to evaluate potential control treatments

Compliance with statutory regulations

To carry out a field trial in the United Kingdom, it is necessary to comply with criteria laid down by the Health & Safety Executive, Chemicals Regulation Directorate (CRD). An

Administrative Experimental Approval for Research and Development Work was obtained (COP 2011/01180) and the personnel that carried out the trial had obtained certification (PA1 and PA6) in application of pesticides in accordance with the regulations on use of experimental pesticides.

Simulation of the farm's spray regime

Sprayers used at Intercrop Ltd. are fitted with flat Lurmark Drift-beta nozzles in front of flat fan nozzles. Both apply spray simultaneously at a total rate of 400 litres per Ha. For the experimental spray treatments, compression sprayers (Hozelock 5 litre Killaspray) were used to apply the sprays. The volume for experimental spray plots to mimic the farm application rate was calculated as: $400 * 10/10000$ litres = 0.4 litres per plot.

Sprayers (one was used to apply each treatment) were calibrated as follows. The sprayers were set to give a flow rate of 400 ml per minute. This was achieved by pumping 50 times after 1600 ml of spray liquid had been put in the sprayer. Although only 1200 ml (400 ml x 3) was needed for the trial, dead volume in the sprayer (spray liquid remaining after the spray became intermittent) meant that an additional 400 ml was required in the container (making a total of 1600 ml of spray liquid for each treatment per spray date).

Spray protocol to ensure a precise insecticide dose

Compression sprayers are pumped up before use. In this case, 50 compressions (pumped 50 times) gave the desired flow rate for treating a plot. When liquid is emptied from the reservoir during spraying, the pressure falls and the flow rate is reduced. Laboratory tests prior to the fieldwork had showed that this would have affected the dose applied to subsequent plots. To compensate for the fall in pressure after one minute of spraying (spraying a single plot) it was found that the sprayer needed to have seven additional pumps before the next minute of spraying to reinstate the original pressure. After the second plot had been sprayed another seven pumps were needed. In this way the same flow rate of 400 ml per minute and spray quality was retained for each of the three replicates.

Spray application procedures

There were three replicates for each treatment. Total spray liquid applied to plots for each spray treatment was $3 * 0.4 = 1.2$ litres. However, to allow for the dead volume in the bottom of the spray tank we mixed 1.6 litres. Surplus spray liquid was applied to an adjacent grassed area of the farm.

When treating the plots, the operator sprayed from the side of the plot to avoid walking on the bed. The spraying operation was rehearsed several times with water to practice achieving an even coverage at the required volume rate. The compression sprayers (one for each chemical) had been set to give 400 ml/min flow rate before the trial, so each plot had to be sprayed for 60 seconds. To help pace the operator, ten second intervals were called out by a colleague.

The flow rate of 400 ml/min was set by putting into the sprayer 1600 ml of spray liquid, then pumping 50 times before spraying the first plot. After completing a plot the sprayer was then pumped a further seven times before spraying the next plot to maintain pressure and flow rate.

The plots required small quantities of the supplied pesticide formulations, so these quantities were measured by weight on a four figure balance. All samples were pre-prepared and held in sealed, labelled glass bottles before use (Table 1).

Table 1. The experimental insecticide rates for plots in the 2011 leafminer trial.

Treat-ment No.	Product and recommended dose rate	Quantity (units) per plot	Quantity (units) for three plots	Quantity per vial allowing for dead spray in tank	No. of vials required (incl. one extra)
		400 ml	1200 ml	1600 ml	
1	Control	0	0	0	0
2	Deltamethrin Decis Protech 1.5% Farm rate is 420ml/ha (could have a max of 4 treatments per crop)	0.42 ml	1.26 ml	1.68 ml	4 vials
3	Acetamaprid (Gazelle) SG 0.25 kg/ha (2 sprays per crop and at least 7 days before harvest)	0.25 g	0.75 g	1.0 g	3 vials
4	<i>B.thuringiensis</i> var <i>kurstaki</i> (DiPel DF) – reduced rate of 0.5 kg/ha (max dose of 1 kg /ha)	0.5 g	1.5 g	2.0 g	3 vials
5	Spinosad (Tracer 480 g/l) Use at 200 ml/ha (max of 3 treatments per crop)	0.2 ml	0.6 ml	0.8 ml	4 vials
6	Spirotetramat (Movento) 0.5 l/ha (two treatments per crop)	0.5 ml	1.5 ml	2.0 ml	3 vials

Treat-ment No.	Product and recommended dose rate	Quantity (units) per plot	Quantity (units) for three plots	Quantity per vial allowing for dead spray in tank	No. of vials required (incl. one extra)
	<i>Volume of spray</i>	400 ml	1200 ml	1600 ml	
7	*Coded product. HDCI 015 10% a.i. 0.5 l/ha (two treatments per crop)	0.5 ml	1.5 ml	2.0 ml	3 vials
8	Chlorantraniliprole (Coragen 20SC) 175 ml/ha (two treatments per crop)	0.175 ml	0.525 ml	0.7 ml	3 vials
9	Diflubenzuron (Dimilin Flo) 0.2 l/ha (two treatments per crop)	0.2 ml	0.6 ml	0.8 ml	3 vials
10	Orophite 0.5% concentration (two treatments per crop)	2 ml	6 ml	8 ml	3 vials
11	*Coded product. HDCI 015 10% a.i. 0.3 l/ha (two treatments per crop)	0.3 ml	0.9 ml	1.2 ml	3 vials
12	Abamectin (Dynamec)**	0.2 ml	0.6 ml	0.8 ml	1 vial

*DPX-HGW86 100 – persistence is 7-10 days.

**This active was included after the start of the trial and so only one spray could be applied.

The trial site

The trial was carried out on the Greenacres field of Intercrop Ltd. Farm, Kent. To fit in with the farm's watering schedule the Farm Manager provided three adjacent beds along an approximately 100 m length of the field. The experimental area also included an untreated and blank 'guard' row at each side of the layout. An unsprayed area of exposed soil (4 m in length) separated the plots along each row.

The trial treatments

Field trial data from a previous project (FV 301) showed that systemic insecticide seed treatments (e.g. thiamethoxam) did reduce mining damage in tatsoi leaves. Actives that exhibit systemic and translaminar efficacy, therefore, were considered to have the most potential for managing leafminer outbreaks, especially when these might initially have gone unnoticed.

A search of the HSE CRD pesticides databases did not produce any results for insecticidal treatments aimed specifically at leafminers on outdoor chard or spinach. A wider search,

therefore, of the currently recommended insecticides for use against leafminers was carried out and the following candidate treatments were selected for the field trial.

- I. Untreated control with no leaf miners released (required for proper statistical analysis of the data).
- II. Deltamethrin spray (Bayer Decis Protech). A type of 'control' treatment, which may provide protection against adult leaf miner oviposition (egg laying). It is one of the standard actives already used by leafy salad growers (SOLA for outdoor spinach).
- III. Acetamaprid (Gazelle) as a spray treatment. To provide protection during the early stage of the crop. Acetamaprid had a SOLA for use on outdoor spinach.
- IV. *Bacillus thuringiensis var kurstaki* (DiPel DF). This treatment would be aimed at the first instar larvae, which need to ingest this active. It has a SOLA for outdoor spinach until 2013.
- V. Spinosad (Dow Tracer 480 SC). Has been used to control leaf miners on other crops and has no significant effect on beneficials (has a SOLA until 2017 for chard and spinach).
- VI. Spirotetramat (Movento) This active exhibits systemic and translaminar efficacy, whereas its contact efficacy is rather limited. It could be highly effective at preventing maggots from developing in leaves.
- VII. Coded product (HDCI 015) – is reported in the USA to have good activity against Diptera leafminers. This treatment was recommended to be carried out at two rates (high and low, see Table 1).
- VIII. Diflubenzuron (Dimilin Flo). This active has been reported to suppress leaf miner populations and does not affect beneficial insects significantly. Diflubenzuron has a SOLA for outdoor spinach until 2013.

In addition to those actives listed above, and after discussion with the Industry Representatives, Coragen (chlorantraniliprole), Orophite (a proprietary supplemental foliar feed with a strong orange smell) and Dynamec (abamectin) were included as additional treatments (Table 1).

The experimental trial design was randomised with three replicates of each treatment. Treatments were applied as per the schedule below (Table 2) As no previous trial data were available to 'benchmark' the optimal timing or number of sprays, it was decided after discussion with the Industry Representative to apply a third treatment to a randomly selected half of each of the deltamethrin and spinosad treatment plots. The primary reason for doing this was that the trial relied on pest pressure from local leafminer populations

(rather than from caged insects, as planned initially) and so its duration was extended by approximately one week longer than the normal harvest time. This increased the likelihood that the trial would experience significant leafminer pressure, as well as ensure that those actives with a definite effect on leafminers could be identified clearly.

Table 2. Leafminer trial spraying schedule for 2011

Dates	Crop treatments and assessments	Spray treatments applied
Day 1 Thu 18 August 2011	<ul style="list-style-type: none"> ➤ Red Chard Crop drilled. ➤ Plots irrigated. ➤ Blend 000524.5 and Granular Urea (fertilizer) applied. ➤ Delicia slug- lentils applied. ➤ Maxicrop Triple (plant growth stimulant). ➤ Pyramin (soil acting herbicide). 	No
Day 7 Thu 25 August 2011	<ul style="list-style-type: none"> ➤ Crop emerged, but about seven days from true leaf emergence. ➤ Large numbers of <i>P. hyoscyami</i> eggs on volunteer plants next to the main path. Also, an adult leaf-miner caught on the plants. ➤ Plots allocated to treatments. ➤ <i>P. hyoscyami</i> adults beginning to emerge from mass-reared pupae and so well timed for the trial. 	No
Day 13 Wed 31 August 2011	<ul style="list-style-type: none"> ➤ Crop assessed for spraying. 	No
Day 14 Thu 1 Sept.2011	<ul style="list-style-type: none"> ➤ Crop sprayed for first time. All treatments sprayed. ➤ 'Bioassay' plants sprayed. 	Yes
Day 21 Thu 8 Sept 2011	<ul style="list-style-type: none"> ➤ Crop sprayed for second time. All treatments sprayed. 	Yes
Day 28 Thu 15 Sept 2011	<ul style="list-style-type: none"> ➤ Only deltamethrin and spinosad treatments sprayed for a third time (randomly selected halves of the plots). 	Yes
Day 35 Thu 22 Sept 2011	<ul style="list-style-type: none"> ➤ Final damage assessments. 	No

Damage data were collected by dividing each plot into 40 equally sized sub-plots. Each of these sub-plots was then examined carefully for the presence of mines and the number of mines per sub-plot recorded. These data were analysed by analyses of variance using the statistical package GenStat.

At the proposal planning stage, it was hoped to have leafminer adults present in cages positioned over the trial plots to ensure adequate pest pressure. Preliminary tests of this concept revealed difficulties with this plan, however, due to the basic biology of the

leafminer species, *P. hyoscyami*, responsible for the problem. Adult females laid a relatively small number of large eggs, compared to other pest species, such as noctuid moths, which can produce several hundred eggs per female. Also, when placed in a cage in an open field, even with sources of honey solution available, adult-fly mortality was very high and oviposition low.

The low 'pest pressure' this would have generated meant that the original plan was impractical and that a better use of the adults was to carry out a small 'bioassay' test that involved spraying plants (3 - 5 plants per insecticide treatment) on which leafminer populations were developing. In order to achieve this, oviposition was allowed to occur on the test plants for approximately one week, after which the plants were transported to the field trial site where they were treated once with the appropriate active, as part of the spraying regime (on day 14). After spraying, the plants were allowed to dry and individual leaves with eggs or mines numbered, tagged and photographed. Plants were then covered individually with fresh perforated, plastic bread bags and left for two weeks to allow any damage to develop. At this point they were assessed by comparing the images of the tagged leaves with their current condition. Leaves that had continued to experience damage after spraying were rated "1", whereas those leaves where mining had ceased after spraying were rated "0".

Results

Collection of leafminer species

Adult flies were collected by sweep-net in April-May, both from Intercrop Ltd and from allotments in the Medway area. Most of the flies collected using this technique were not chard or spinach pests, although a few *P. hyoscyami* adults were caught. A more productive method of collecting chard and spinach leafminers proved to be digging up the soil beneath mined plants. By mid-May (14th – 21st), the first generation of leafminers had reached the pupal stage and large numbers of *P. hyoscyami* pupae were collected beneath mined, volunteer, red chard and fat hen plants (Figure 5). The adults that emerged from these pupae were used to initiate the colony for mass rearing (Figure 6). Project work began in April 2011 and so it is assumed that the overwintering population that gave rise to these pupae must have emerged in mid- to late March.



Figure 5. *P. hyoscyami* pupae were collected beneath mined, volunteer, red chard (LHS) and fat hen plants (RHS) in May 2011.



Figure 6. An adult female *P. hyoscyami* that emerged from the field-collected pupae. These adults were used to initiate the colony for mass rearing.

In order to assess pest pressure throughout the summer, small 'trap' plants of red chard were planted at bi-monthly intervals over the summer and taken to Intercrop Ltd. and to an organic allotment in Medway (Figure 7).



Figure 7. An example of the ‘trap’ plants used to collect eggs of chard and spinach leafminer species.

The trap plants were monitored for eggs and adult female *P. hyoscyami* could often be seen in the late afternoon ovipositing on them. From June onwards, there was a consistent amount of low pest pressure from *P. hyoscyami* and new eggs were found on the trap plants in the organic allotment on most days.

From mid-September onwards, oviposition on the trap plants ceased and this coincided with this generation of *P. hyoscyami* larvae in the colony entering a pupal diapause.

In April – May, blue sticky traps (i.e. an attractive colour to Diptera) were placed at the edges of chard and spinach crops to sample the adult-fly population. These caught a very large number of fly species in a short time (three to four days) (Figure 8). Many of these species were not pests, but to the untrained eye would be hard to differentiate from *P. hyoscyami* (Figure 9). It was thought that this monitoring method, therefore, would be of limited use to growers and so this activity was not continued throughout the summer.

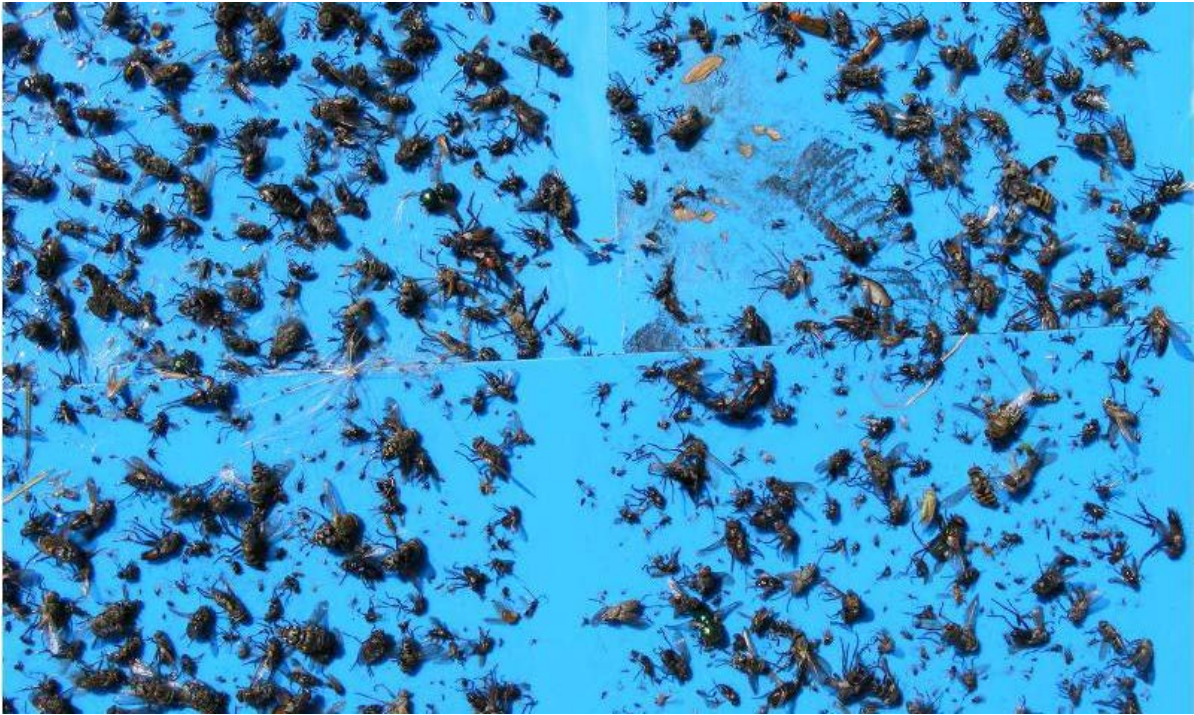


Figure 8. Examples of the large number for fly species that were caught on blue sticky traps at Intercrop Ltd, during a three to four day period in early summer.



Figure 9. A *P. hyoscyami* female that, to the non-entomologist, would look much like many other fly species.

In order to assess leafminer pressure in another area of the country and to collect samples, a visit was made to the Emmett (Nottinghamshire) farm (NG22 8TW) in late summer (20 September 2011), around the same time as the start of the field trial at intercrop Ltd. The Emmett farm manager said that they had not noticed any leafminer problem and a survey of the crops and surrounding area did not reveal any mined plants. This suggests that as well as being sporadic in nature, this pest species may also have local hot-spots.

Mass rearing of leafminers

The large number of pupae collected in May (>200), were used to initiate the mass rearing colony. The colony was established successfully and large numbers of larvae reared to provide the first generation of mass-reared flies in late June (Figure 6).



Figure 10. Plants with a large number of *P. hyoscyami* mines that were used to mass rear this species.

Due to the relatively long egg-to-egg life-span of seven to eight weeks, only one intervening generation was then available before adults flies were required for the field trial, which was planned for late August – September. Some adults from the June emergence, therefore, were used to test how efficiently they could provide pest pressure while confined in cages positioned over red chard. The results of this pre-trial were disappointing in that the mortality of the adult flies was very high and oviposition low. It is probable that the reason for this was because the artificial caged conditions were quite harsh and did not provide the flies with an opportunity to conduct their normal courtship and mating behaviours. In the light of this pre-trial, it was decided to use the mass reared adult flies in an alternative and potentially more productive bioassay experiment.

Field trial to evaluate potential control treatments

The field trial was timed successfully to catch the late-summer emergence and oviposition by *P. hyoscyami* adults. Damage from mining was clearly evident in the control plots and the presence of a significant number of larvae in the leaves would have caused the crop to have been rejected (Figure 11).



Figure 11. A *P. hyoscyami* larva mining a leaf in one of the control plots. Larvae were often observed leaving the mines to move across leaves, particularly during the early morning. Exit holes from the mine can be seen on the right hand side of the image.

The third sprays of deltamethrin and spinosad did not affect damage significantly and so data for both halves of these plots were pooled. Several of the actives prevented the leafminer damage to an impressive degree. Deltamethrin was the most effective, followed by the coded product applied at the higher rate. Acetamaprid also provided good protection and where mines were found in these treatments, it was usually at the edges of the plots where spray coverage may not have been applied so evenly.

In the field, diflubenzuron reduced the amount of damage compared to the untreated control, but this result was inconsistent with the 'bioassay' data, where damage to leaves continued after spraying (see Figure 16 below). Chlorantraniliprole also reduced damage, but many large mines were found in the plots and it was felt that this level of damage would be unacceptable to the growers and consumers.

The spirotetramat, abamectin, spinosad, *B. thuringiensis*, and Orophite treatments all experienced damage levels similar to that of the control plots (Figure 12).

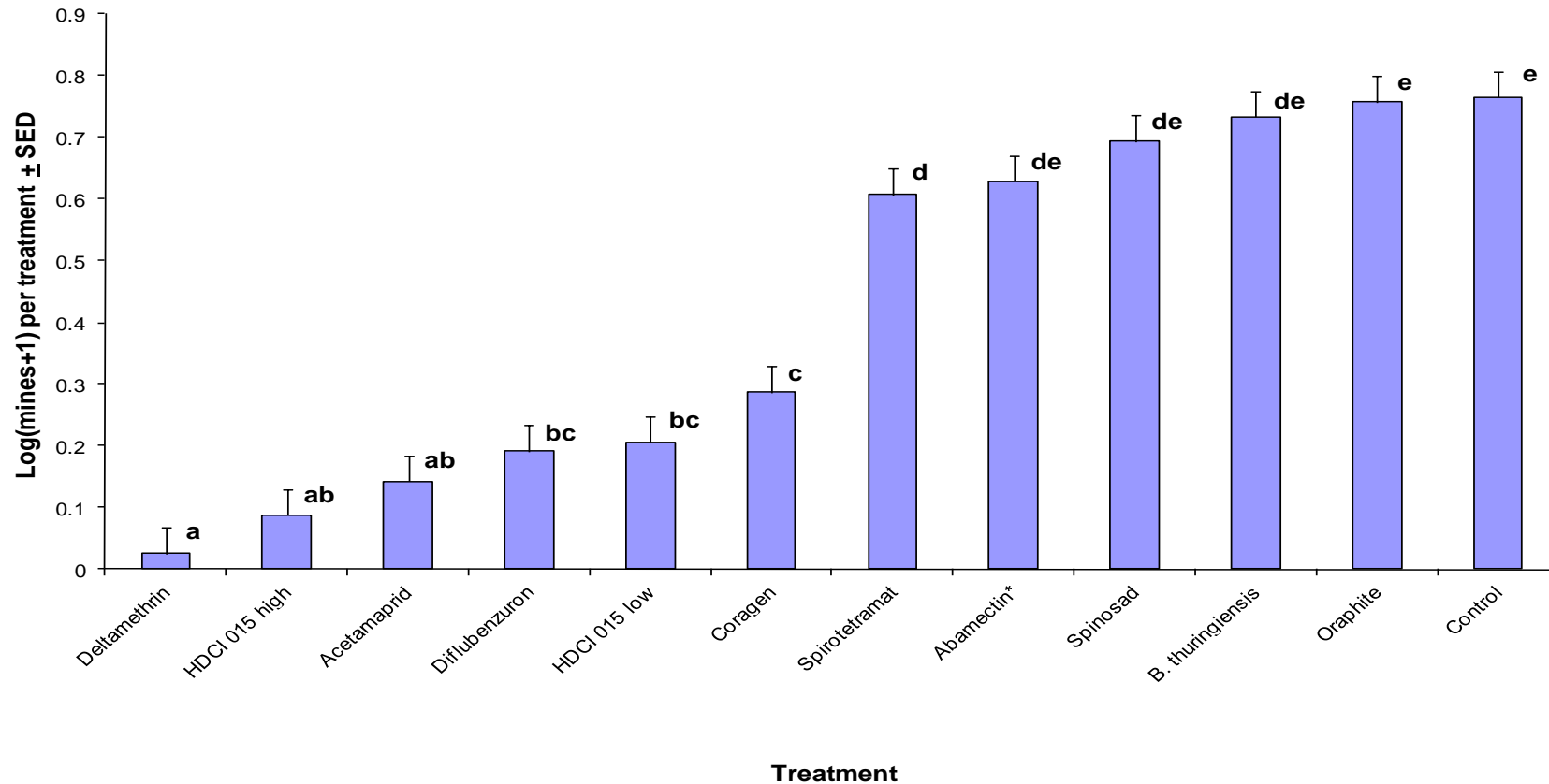


Figure 12. Chard and spinach leafminer trial (18 August – 22 September 2011). The mean numbers of leafminer mines present in the different experimental treatments of a red chard field trial on 22 September 2011, four weeks after sowing. Damage data were collected by dividing plots into 20 equal areas and counting the number of mines in each of the 20 areas. These data were $\text{Log}_{10}(x+1)$ transformed and a mean calculated per plot. ANOVA was carried out on transformed data, followed by Tukey's pairwise comparisons at the $P < 0.05$ significance level. Means with the same adjacent letters are not significantly different. Error bars are standard errors of differences of means. *The abamectin treatment consisted of one spray only on 15th September 2011.

Spraying of plants infested artificially with P. hyoscyami ('bioassay experiment')

This small bioassay trial produced data similar to that of the field trial. Deltamethrin, the coded product at both rates and acetamaprid all prevented further damage to plants after spraying and had damage score totals of zero or 1 (Figures 13 - 15). Diflubenzuron, however, and the other actives did not prevent further damage (Figure 16) and in most cases the leafminer larvae developed successfully into pupae. This group of actives all had much higher total damage scores of three to five. As the actives fell into two clear-cut groups, i.e. no further damage, or continuing damage, a statistical analysis was unnecessary.



Figure 13. An example bioassay plant that was sprayed with deltamethrin. Even though eggs and mines were present before spraying, damage ceased after the plant was sprayed.



Figure 14. The same leaf before and two weeks after spraying with the coded product (HDCI 015). The identical shape of the mine indicates that this active had caused the almost immediate death of the leafminer larvae.



Figure 15. The effect of the acetamaprid spray in preventing further damage to the experimental plant. A dead larva can be seen in the bottom of the mine in the RHS image.



Figure 16. Although the diflubenzuron spray reduced leafminer damage in the field trial, it was ineffective on the experimental bioassay plants and mining damage was very severe two weeks after spraying.

Discussion

The data presented in this report are from a single field season and so the conclusions that can be drawn need to be done so with this proviso in mind. In 2011, the only leafminer pest species attacking chard and spinach crops of any significance was the beet leafminer or

mangold fly, *P. hyoscyami*. Other insect species such as aphids and silver-Y moth caterpillars were seen, but the damage caused by them was very limited.

Various methods were used to sample and collect leafminers associated with chard and spinach crops. One particularly effective method was to collect the pupae from the soil underneath mined plants early in the year (May). The adults that emerged from these pupae provide the first real pest pressure of the season and so removal of volunteer chard and spinach plants earlier in the year would be an effective cultural control method for reducing pest pressure.

The insect rearing confirmed that there are probably only three generations of *P. hyoscyami* most years, which to some extent explains the sporadic nature of the problem. Although more detailed monitoring would be required to confirm this, it is expected that leafminer problems are therefore most likely to occur in south Kent during late May to mid June and from mid-August to September.

In order to monitor leafminer pressure, blue sticky traps are unlikely to be the answer due to the large fly populations, most of which were not *P. hyoscyami*. A much more effective monitoring method would be to plant a small area of 'trap' red chard at monthly intervals, adjacent to the chard and spinach growing fields. These plants could be monitored quickly and efficiently for presence of the distinctive white eggs. When new eggs were found, this would indicate that scouting the main crops would be necessary, followed by insecticide sprays, if appropriate.

The field trial showed clearly that deltamethrin, the coded product (HDCI 015) and acetamaprid all reduced leafminer damage substantially and prevented larvae maturing to become large white maggots. The effect achieved by deltamethrin was unexpected, mainly because synthetic pyrethroids are not reported to have any systemic activity within plants. It is likely, however, that its activity was achieved by an ovicidal effect, as well as knock-down or irritant effects on adult flies visiting the crop. Either of these modes of action would reduce oviposition by adult females and the subsequent survival of any eggs they managed to lay. The behaviours of *P. hyoscyami* larvae may also contribute to deltamethrin's efficacy. Larvae were observed to leave their mines, particularly at night and during the early morning. When this occurs, these behaviours will increase the likelihood of picking up a lethal dose of deltamethrin, while crawling over the sprayed leaf surfaces.

The coded product and acetamaprid both showed excellent activity against *P. hyoscyami*. These products are particularly useful against this pest, because if for any reason an

outbreak of the pest is missed during the early stage of infestation, use of these products will ensure that no large larvae develop in the crop.

The small experimental bioassay experiment produced clear-cut data that supported the field-trial results. The only active where there was a marked discrepancy between the field and bioassay data was diflubenzuron. One reason for this may have been that many of the larvae had already entered the plant in the bioassay and so were in a protected environment at the time of spraying. In the field, the first spray was applied early in the life of the crop and probably prior to much of the oviposition that occurred. Under the field-trial conditions, therefore, younger larvae would have picked up a dose of diflubenzuron while entering the leaf and so would have suffered a much higher mortality rate. None of the other actives provided sufficient leafminer control, either in the field or the bioassay.

The data presented above provide clear information on which to base leafminer management practices. The precise modes of action of the different control options remain speculative at the moment and determining them would require additional careful bioassay work. It is possible to say, however, that there is no indication at present that the *P. hyoscyami* population at Intercrop Ltd. has developed resistance to any of the insecticides. The other very promising finding is that each of the three most effective actives have different modes of action. This will help reduce the risk of resistance development and ensure the longer-term sustainable production of spinach and chard crops, even in *P. hyoscyami* 'hot-spots'.

Conclusions

- In 2011, the only leafminer pest species attacking chard and spinach crops of any significance was the beet leafminer or mangold fly, *P. hyoscyami*.
- Removal of volunteer chard and spinach plants earlier in the year would be an effective cultural control method for reducing pest pressure.
- There are probably only three generations of *P. hyoscyami* in most years. Egg laying by leafminers is therefore most likely to occur in south Kent during late May to mid-June and from mid-August to September.

- An effective and direct monitoring method would be to plant a small area of 'trap' red chard at monthly intervals and check these regularly for the distinctive white eggs.
- Of the actives tested, deltamethrin, the coded product and acetamaprid all reduced leafminer damage substantially and prevented larvae maturing to become large white maggots.
- No evidence of resistance development was detected.
- The effective actives all have different modes of action, which will reduce the risk of resistance development and thus ensure the longer-term sustainable production of spinach and chard crops, even in *P. hyoscyami* 'hot-spots'.

Knowledge and Technology Transfer

- A presentation was made to the SPGA panel.
- Photographic images and text were supplied to Ms Grace Choto for an article in the British Leafy Salads Association Newsletter.
- An article is planned for the HDC News.