Project title:	Biology of the cabbage whitefly, *Aleyrodes proletella*
Project number:	CP 091
Project leader:	Dr Rosemary Collier
Report:	Annual report, October 2014
Previous report:	N/A
Key staff:	Spencer Collins (PhD student)
Location of project:	Warwick Crop Centre School of Life Sciences University of Warwick Wellesbourne CV35 9EF
Industry Representative:	Andy Blair, Emmetts, UK.
Date project commenced:	01 October 2012
Date project completed (or expected completion date):	31 December 2015 (thesis completion)

DISCLAIMER

[The results and conclusions in this report are based on an investigation conducted over a one-year period. The conditions under which the experiments were carried out and the results have been reported in detail and with accuracy. However, because of the biological nature of the work it must be borne in mind that different circumstances and conditions could produce different results. Therefore, care must be taken with interpretation of the results, especially if they are used as the basis for commercial product recommendations.]

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.

[Name]
[Position]
[Organisation]

Signature .... Date ....

[Name]
[Position]
[Organisation]

Signature .... Date ....

Report authorised by:

[Name]
[Position]
[Organisation]

Signature .... Date ....

[Name]
[Position]
[Organisation]

Signature .... Date ....

CONTENTS

GROWER SUMMARY

Headline

Field sampling, plot trials and laboratory experiments are providing information that is leading to an improved understanding of cabbage whitefly ecology**.**

Background

The cabbage whitefly, *Aleyrodes proletella,* has become an increasing problem for the Brassica industry in recent years, especially on Brussels sprout and kale. The reason for this is unknown, but it is believed to be due to a combination of climate change, removal of certain active ingredients from use and later harvest times of crops. Little research has focused on this species as, historically, it has been regarded as a minor pest. Knowledge about the biology of the cabbage whitefly is limited and most of what is currently understood about its ecology has been inferred from minimal anecdotal evidence.

The overall aim of this project is to understand population trends of *Aleyrodes proletella* in the most vulnerable crops, Brussels sprout and kale. This includes understanding the key times of population/generation increase and colonisation of a new crop.

Studies are focusing on periods of colonisation of crops, natural population increases within the field and the relationship between development rate and temperature. These investigations will build on previous knowledge and lead to the construction of a phenological model to help farmers develop targeted control methods that will maximize efficacy. An understanding of the weather conditions that impact on survivorship will also lead to an ability to forecast years that would support large numbers of whitefly, allowing growers to focus only on periods when treatments are absolutely necessary. Potential overwintering locations that may act as reservoir sources for the pest will be investigated, together with the dispersal potential of the species. It is hoped that through empirical study of this species we will gain an increased understanding of its ecology which can lead to an integrated management scheme to both prevent the development of large infestations within a crop and increase the efficacy of treatments applied to reduce whitefly infestations.

Summary

Experiment 1. Monitoring of whitefly on a newly-planted vulnerable field crop (Brussels sprout and kale) throughout the season.

Plots of Brussels sprout and kale were planted to investigate natural colonisation and population increase of the cabbage whitefly over a season. Plots consisted of 24 plants (3x8, 50cm spacing) of both kale (cv. Reflex) and Brussels sprout (cv. Revenge) (see Figure 1). Five replicated plots were planted in different locations on the Wellesbourne site. Plants were sown in 2x2cm modules and raised in glasshouse conditions for 7 weeks prior to planting out in the field. To determine date of colonisation to within a week, all plants and all leaves of all plots were surveyed weekly for a month after planting.

Kale and Brussels sprout plants supported differing numbers of whitefly of all life stages. However, this difference was only apparent after August, with the greatest difference occurring during the winter months. The presence of similar numbers of adults for the first few months suggests that there was no difference in the initial attractiveness of the two crops. In addition, the similarity in egg numbers at first suggests that, in the case of immigrating females, host plant quality was similar. This supports the results of laboratory work on egg-laying rate on kale and Brussels sprout plants which showed no significant difference between the two crops (Year 1 report, Experiment 1.4 (Collins, 2013)). As differences became more apparent after a full generation had developed on the plants, it is possible that females developing on the different crops emerge with different fecundities and future work will investigate this further. The extra physical protection provided by kale, in terms of plant structure, when compared to Brussels sprout plants may also have an effect. Adult whitefly may be less likely to be dislodged from kale by rain and wind. Future experiments will investigate whether this assumption is a valid one. When considering the crops separately, the 5 plots supported similar numbers of whitefly, with significant differences occurring on just a few occasions.

Experiment 2. Height at which adult whitefly disperse.

Using a telescopic flagpole, sticky traps were deployed at various heights from the ground (ground-420cm, 60cm intervals) to monitor adult whitefly and determine the vertical distribution of flight. The sticky traps (22cm x 10cm) were either yellow or blue (BHGS Ltd, UK) and were rolled into a cylinder (10cm tall, 8cm diameter) to provide a coloured sticky surface that covered 360°. After deployment for 7 days, the traps were collected and the numbers of whitefly counted using a microscope. The trapping periods began on 28th May, 14th June and 28th June 2014. Yellow sticky traps at ground level caught significantly more adult whitefly than any other trap colour/height combination, catching on average ~50 individuals per trap during the study period.

The next highest catch rate was by yellow traps at 60cm and here the average catch rate fell to only ~5 individuals per trap. Yellow traps caught significantly more whitefly than blue traps but this was only apparent up to 120cm, when numbers captured were too low to distinguish between them. These results show the importance of trap height in terms of the efficacy of yellow sticky traps for monitoring the cabbage whitefly. A difference of 60cm can reduce catch rates of whitefly by ten-fold. Any sticky traps used for monitoring cabbage whitefly are likely be much more effective at ground level. Data shown in this report only represents the behaviour of reproductively-active whitefly. However, previous studies have suggested that female cabbage whitefly entering reproductive diapause show stronger migratory behaviours (Iheagwam, 1977b). The current study will be replicated at times when females in diapause are present; during September and October, for example.

Experiment 3. Distribution of whitefly in a commercial crop of kale.

Adult whitefly were counted on individual plants within a commercially-grown field of kale, cv. Reflex, in Lincolnshire. Transects were taken from each side of the four field edges (North, South, East and West). Whitefly were counted at distances of 0 (edge plants), 5, 15, 35, 75 and 155 plants into the field, plant spacing was ~60cm. Four plants at each distance were sampled for whitefly using a similar approach to Experiment 1. As the numbers of whitefly in each transect seemed to differ, each field edge was sampled in more detail. Plants were sampled in five randomised locations on each field edge. Sampling was conducted on 18th August 2014. Average numbers of whitefly on plants at the edge were approximately 4 times greater than those at a distance of 45m into the field, showing a clear edge effect in numbers of cabbage whitefly. Comparing numbers of adults between each of the field edges (North, South, East and West) showed that numbers of adult whitefly differed between them. The most pronounced difference was between the northern and southern edges where there was an approximately five-fold difference in numbers of adult whitefly. This information is very important for growers who are surveying fields for cabbage whitefly. If counts are always made from the same edge of the field, growers are unlikely to get a true representation of the field as a whole. Further research will be needed on multiple fields and at multiple times of year, to see if this type of distribution applies to all field situations.

Experiment 4. Estimating the abundance of adult whitefly at different distances from a field infestation.

Yellow sticky traps and ‘trap plants’ were deployed in ‘transects’ at a range of distances from plots infested with whitefly, which were those used in Experiment 1. Each sticky trap was placed horizontally on a plastic tray ~1 cm above the ground. For the approach using ‘trap plants’, groups of three cauliflower plants (7^{th} true leaf stage) were placed at the required distances along the hedge line adjacent to the infested plot. Both the sticky traps and trap plants were left for 7 days, after which all adults on the plants were counted by eye. The sticky traps were taken into the laboratory where the whitefly were counted using a microscope. The sticky trap method was used in each of 3 locations on 3^{rd} and 30^{th} April, 2014. The trap plant method was used in each of 3 locations on 24^{th} April and 9^{th} June 2014.

Trap plants

There was a significant decrease in the number of adult whitefly found between 0 and 5 metres from the plots, but the numbers found on plants placed at distances of 5 and 15 metres did not differ. At 25 metres, the number of whitely was nearly 5 times less than that at 5 metres.

Yellow sticky traps

The number of whitefly caught on yellow sticky traps at distances of 0 and 5 metres from the population source differed significantly from each other and from all other traps within the transect. The numbers captured on traps located at the remaining distances of 10m, 20m and 30m caught very similar numbers of whitefly and did not catch significantly different numbers from each other.

Experiment 5. Monitoring whitefly activity using yellow sticky traps on the ground.

Yellow sticky traps were placed horizontally and 1cm above the ground on the North, South, East and West sides of the plots used in Experiment 1. Traps were deployed for a 7 day period prior to a sampling event within the plots. The whitefly on the traps were counted using a microscope. Weather data were collected for all days when traps were present in the field. To reduce the potential for dead whitefly to fall onto the traps, they were placed 30cm from the nearest plant, since the aim was to capture actively flying individuals. An ‘activity index’ was calculated, defined as the logged number of whitefly caught/mean number of adult whitefly per plant on the plot. An increase in activity in October by overwintering females supported the hypothesis that they are likely to show stronger migratory behaviour. As expected, the coldest months of December and January were also those showing the lowest levels of activity. Activity of overwintering females increased through March until May. A sharp increase in activity at the time of the emergence of the first generation in May-June supports the notion that first generation adults are likely to move onto new hosts and it signals a time when new crops are likely to be colonised. For the majority of the year, the direction in which the sticky trap was placed (i.e. northern, southern, eastern and western sides of the plot) had no effect on the catch rate. However, during December and January more whiteflies were captured on the traps placed to the North and the East. This also corresponds to the period when the whitefly had the lowest activity index. It is likely that trap captures at this time of year are whitefly that have been dislodged from their host rather than actively leaving the crop. The wind direction at Wellesbourne is predominantly south-westerly, this would cause any whitefly that are dislodged to be blown towards the northern and eastern traps more often.

Although a significant relationship between the numbers of whitefly caught on sticky traps and the numbers of whitefly per plant (both in log numbers) using sticky trap catches to monitor crop populations is unlikely to be reliable, it is likely that other factors contribute to catch rate, such as the reproductive condition of females and the ambient temperature. Using sticky traps to provide an estimate of adult whitefly populations on a crop is unlikely to be accurate and obviously this cannot be used estimate juvenile populations within the crop.

Experiment 6. Monitoring immigration and establishment of whitefly populations on spatially- and temporally- separated plantings of kale.

Plots of Kale (cv. Reflex) were planted in 5 locations at Warwick Crop Centre, Wellesbourne. Each plot consisted of 5 sub-plots of 6x6 kale plants separated by ~18m. A single sub-plot was planted at each location on 19th May, 17th June, 19th July, 15th August and 16th September. Plants were sown in modules and raised in glasshouse conditions for 5 weeks prior to transplanting in the field. For the first month, all leaves of all plants were surveyed for the presence of whitefly. When plant size increased, together with the size of the infestation, 10 randomly-selected plants were sampled completely. This experiment is on-going, and collection of data from all the sub-plots is not complete. Only data from the May plantings is included in this report. Whitefly numbers increased quickly in this study; the mean number of whitefly per plant in plot F exceeded 10 after only 36 days. In contrast, this did not happen in 2013 (in Experiment 1) until after 2 months. As the plots from 2013 (Experiment 1) were still present, there was a larger overwintering whitefly population at Wellesbourne in 2013-14 than in 2012-13. The closer a 2014 plot was to a plot planted in the previous year, the higher the rates of immigration it received. Doubling the distance from the source from 50m to 100m corresponded to a 75% reduction in the numbers of whitefly (after 13 days). This suggests, a very reasonable conclusion, that the rate of colonisation by whitefly onto new crops is highly influenced by the distance of the new crop from sources of overwintering females. Such locations are likely to be overwintered brassica crops such as kale, cauliflower and oilseed rape. This experiment is on-going and, for example, relationships between the colonisation rates and the time of planting will be investigated. Inter-plot differences will also be monitored to ascertain whether the differences observed between the May plantings continue throughout the season.

Financial Benefits

Brussels sprout crops constituted a £51 million 'farm-gate' value for the UK in 2012 (DEFRA, 2012) and the market for kale is increasing. Losses due to whitefly 'damage' are likely to be significant and can lead to product rejections. Brassicas constitute one of the largest users of insecticides within the outdoor-vegetable sector, 22 tonnes of active ingredients used in 2013 (Garthwaite *et al*. 2013). Any techniques that can reduce this insecticide cost will benefit the industry greatly.

Action Points

There are no action points for growers regarding this report.

SCIENCE SECTION

Introduction

Historically the cabbage whitefly (*Aleyrodes proletella*) has been a minor pest of Brassica crops (Butler, 1938a), but recently it has become an increasing pest in Europe particularly of Brussels sprout and kale (Nebrada *et al.* 2005); the cause of which is not fully understood. Effective control with insecticides is difficult due to the adaxial positioning of the nymphs and the leaf structure of the most susceptible crops further adds to the difficulty of achieving good coverage with insecticides. The most effective pesticide for the control of this species is the systemic insecticide Movento® (spirotetramat) (Richter, 2010). Resistance to pyrethroid insecticides has been documented in *A. proletella* (Springate & Colvin, 2011) therefore resistance management is incredibly important. Resistance is often avoided by reduction of pesticide applications. However, sprays must be applied at the most effective times, information that will only be known by studying the biology and life cycle of this pest.

The overall aim of this project is to understand population trends of *Aleyrodes proletella* in the most vulnerable crops, Brussels sprout and kale. This includes understanding the key times of population/generation increase and colonisation of a new crop. This information will be used to suggest periods when application of control methods would be most effective to reduce the overall pest load.

In the scientific literature, whiteflies (Aleyroidae) are a particularly under-represented taxa, including 1556 described species, with potentially many more unknown to science (Martin & Mound, 2007). Work has focused mainly on two species; *Bemisia tabaci* and *Trialeurodes vaporariorum.* This is due to their significant economic importance and very few other species have been studied to anything near their extent. As relatively little is known about the family as a whole, care should be taken not to generalise the findings from these species to all members of the Aleyroidae, as these species are unlikely to be typical of the family (Gerling, 1990).

The basic biology of *A. proletella* was investigated during the 1930's and this gave one of the first insights into this species (Butler, 1938a, Butler, 1938b). The developmental biology of *A. proletella* was investigated subsequently, identifying differences in the low temperature threshold for development in each of the instars (El-Khidir, 1963; Iheagwam, 1978). Although there are suggestions that *A. proletella* can overwinter either as an adult or as a nymph (Iheagwan, 1977a), the overwintering behaviour of the adult females has been the main focus of research. Research has led to an understanding that overwintering is achieved mainly by the ovarian diapause of adult females, induced by shortening day lengths in September (Adams, 1985a; 1985b). The factors leading to the termination of diapause in *A. proletella* have not been elucidated. Chilling has been shown to shorten diapause, and temperatures exceeding 25ºC to prevent diapause (Iheagwam, 1977a). However, this temperature is above that experienced naturally when termination usually occurs (El-Khidir, 1963) and it is likely that a number of factors interact to bring females out of diapause. If a phenological model was to be developed for this species, the factors determining the termination of diapause and the lower threshold of egg laying must be identified.

It is very important to understand the pattern of crop colonisation by *A. proletella*. It has been suggested that overwintering females lay eggs of the first annual generation on their overwintering host. The adults from this generation then migrate to the summer host around June, which is when the first colonisation of a new Brassica crop occurs (Butler, 1938a; El-Khidir, 1963; Al-Houty, 1979). Being able to predict peaks of this migration by developing a phenological model would be invaluable to the Brassica industry allowing a targeted timing of control to eradicate the first colonisers to a new crop.

Materials and methods

Study Site

Unless otherwise stated all field studies were conducted at, or near, Warwick Crop Centre, Wellesbourne, England, CV35 9EF (UK Grid reference SP 27320 56936).

Experiment 1. Monitoring whitefly on newly-planted vulnerable field crops (Brussels sprout and kale) throughout season.

Plots of Brussels sprout and kale were planted on 2nd May 2013 to investigate natural colonisation and population increase of the cabbage whitefly over a season. Plants were sown in modules and raised in glasshouse conditions for 7 weeks prior to transplanting in the field. Plots consisted of 24 plants (3 x 8, 50cm spacing) of both kale (Reflex) and Brussels sprout (Revenge) (Figure 1). Five replicate plots were planted in different locations on the Wellesbourne site as represented in Figure 2 (A-E). Plots were covered in netting to prevent damage by pigeons. To determine the date of first colonisation to within a week, all leaves of all plants were surveyed weekly for a month after planting.

Figure 1. Study plot of Brussels sprout and kale covered in netting to prevent damage by pigeons.

As the sampling effort increased significantly later in the season, due to increased numbers of leaves per plant and numbers of whitefly, a method was developed to optimise sampling. Whitefly have a distinct distribution on plants. Juvenile stages are sessile and age with their host leaves. Adults and eggs are found primarily on young leaves, while pupae are found on older leaves further down the stem (Gould & Naranjo 1999). Such a distribution requires leaves of varied ages to be sampled in order to assess all whitefly life stages without bias. Figure 3 shows a schematic of the sampling approach that was used to assess plants. Analysis of initial data showed that the variance between plants was higher than that within plants, indicating that replication at a plant level would provide better estimations of the population.

Figure 2. Locations of experimental plots for Experiment 1 (A – E) and Experiment 6 (F-J) at Warwick Crop Centre, Wellesbourne.

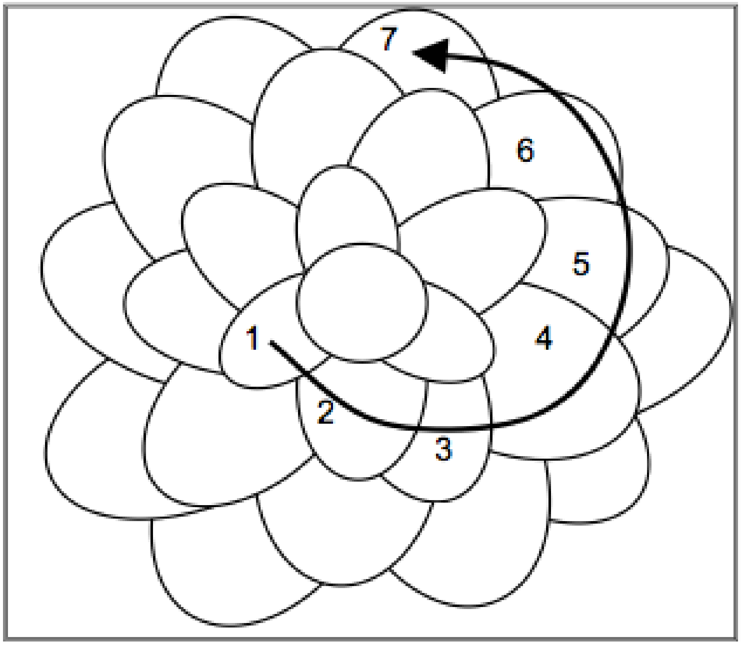

Figure 3. Schematic of sampling plan of plant to incorporate leaf-age distribution of whitefly (From Schultz *et al,* 2010).

Experiment 2. Height at which adult whitefly disperse.

Sticky traps deployed at various heights from the ground were used to monitor adult whitefly to determine the vertical distribution of flight. Telescopic '7 metre Flagpoles' (Newquay camping, UK) were erected within 5m of high cabbage whitefly populations to increase the probability of capture; using the plots in Experiment 1. The apparatus was set up as shown in Figure 4.

The sticky traps (22cm x 10cm) were either yellow or blue (BHGS Ltd, UK) and were rolled into a cylinder (10cm tall, 8cm diameter) to provide a coloured sticky surface that covered 360°. Four replicate flagpoles were set up on the same day and left for 7 days. The traps were then collected and the numbers of whitefly were counted using a microscope, as damaged individuals were easily missed with the naked eye. The trapping periods began on 28th May, 14th June and 28th June 2014.

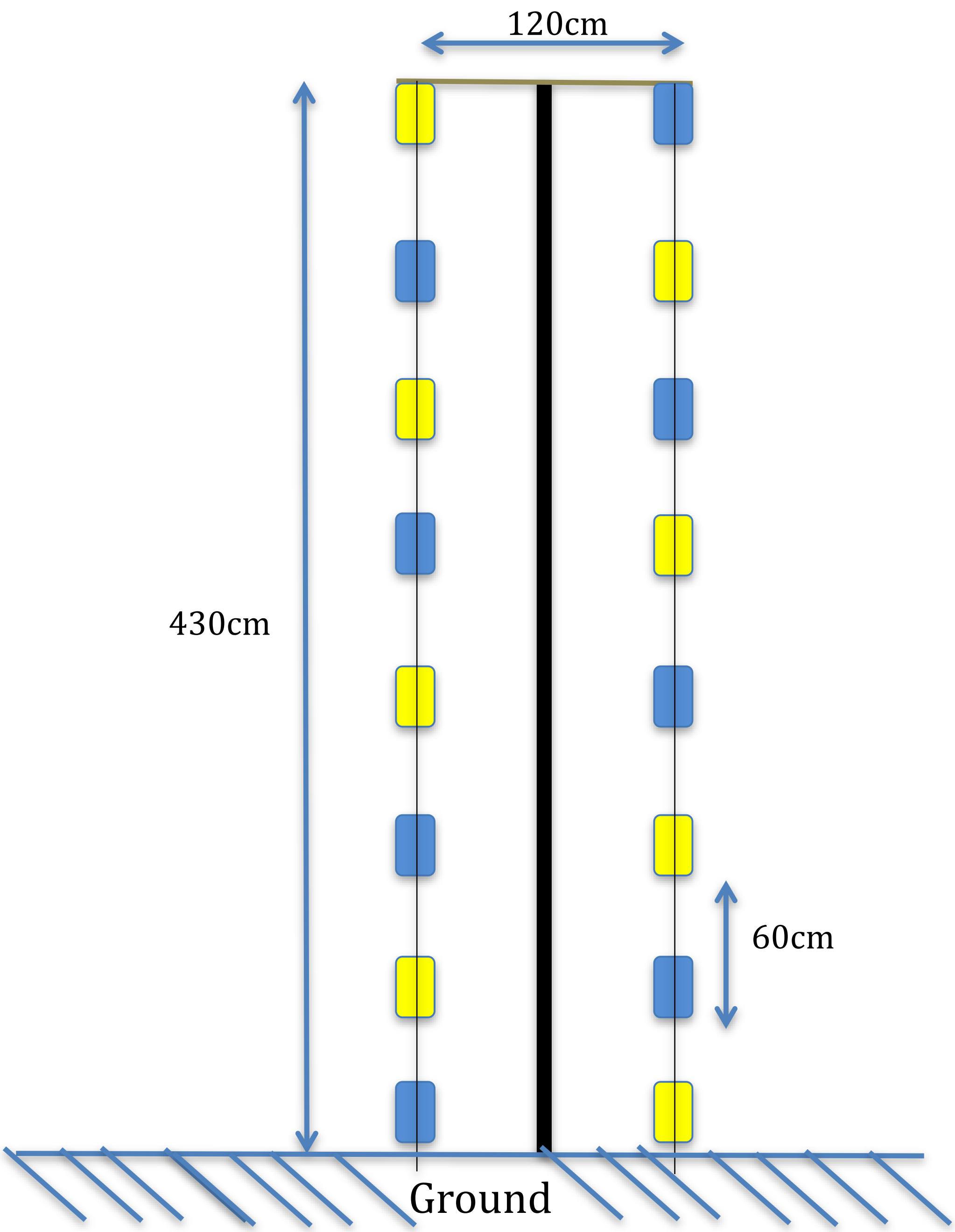

Figure 4. The flagpole supporting yellow and blue sticky traps used in Experiment 2.

Experiment 3. Distribution of whitefly in a commercial crop of kale .

Adult whitefly were counted on individual plants within a commercially grown field (~17 hectares) of kale, cv. Reflex, in Lincolnshire, (Grid reference, TF 33530 21723). Transects were taken from each side of the four field edges (North, South, East and West). Whitefly were counted at distances of 0 (edge plants), 5, 15, 35, 75 and 155 plants into the field, plant spacing was ~60cm. Four plants at each distance were sampled for whitefly using a similar approach to Experiment 1. As the numbers of whitefly in each transect seemed to differ, each field edge was sampled in more detail. The field edges were divided into potential sampling sections of 2 metres and four were selected at random to become replicates for each field edge. The ‘‘0” point of the transect for that edge constituted a fifth replicate. As an edge effect was evident from the transect samples, none of the samples along an edge was taken within 50m of an adjacent field edge. Sampling was conducted on 18th August 2014 and the crop had been in the field for ~3months. The crop had not been treated with insecticide beforehand.

Experiment 4. Estimating the abundance of adult whitefly at different distances from a field infestation.

Yellow sticky traps and ‘trap plants’ were deployed in ‘transects’ at a range of distances from plots infested with whitefly; those used in Experiment 1 (A, D and E). Each sticky trap was placed horizontally on a plastic tray ~1 cm above the ground, this prevented ground active insects (e.g. Carabids) being caught on the trap, which quickly reduced their efficiency to catch other insects. For the approach using ‘trap plants’, groups of three cauliflower plants (7th true leaf stage) were placed at the required distances along the adjacent hedge line of the infested plot. The plants were placed on a 30 x 20cm white plastic tray to hold water and reduce the frequency of watering. Both the sticky trap and trap plants were left for 7 days, after which all adults on the plants were counted by eye. The sticky traps were taken into the laboratory where the whitefly were counted using a microscope. The sticky trap method was used in each of 3 locations beginning on 3rd and 30th April, 2014. The trap plant method was used in each of 3 locations beginning on 24th April and 9th June 2014.

Experiment 5. Monitoring whitefly activity using yellow sticky traps on the ground.

Yellow sticky traps, 22cm x 10cm (BHGS Ltd, UK) were placed horizontally and 1cm above the ground on the North, South, East and West side of the plots used in Experiment 1. Traps were deployed for a 7 day period prior to a sampling event within the plots, so that captures were unaffected by the disturbance caused by assessing the plants in detail. The whitefly on the traps were counted using a microscope. Weather data were collected for all days when traps were present in the field. To reduce the potential for dead whitefly to fall onto the traps, they were placed 30cm from the nearest plant, since the aim was to capture actively flying individuals.

Experiment 6. Monitoring immigration and establishment of whitefly populations on spatially- and temporally- separated plantings of kale.

Plots of Kale (cv. Reflex) were planted in 5 locations at Warwick Crop Centre, Wellesbourne, represented in Figure 2 (F-J). Each plot consisted of 5 sub-plots of 6x6 kale plants separated by ~18m. Figure 5 shows a plan of how each location was set up. A single sub-plot was planted at each location on 19th May, 17th June, 19th July, 15th August and 16th September. Plants were sown in modules and raised in glasshouse conditions for 5 weeks prior to transplanting in the field. Before transplanting the plants were treated with Dursban® (chlorpyrifos) to reduce the risk of damage due to cabbage root fly. For the first month, all leaves of all plants were surveyed for the presence of whitefly. When plant size increased, together with the size of the infestation, 10 randomly-selected plants were sampled completely. However, when the plants consisted of 9 or more leaves, the sampling method shown in Figure 2 was adopted. This experiment is on-going, and collection of data from all the sub-plots is not complete. Only data from the May plantings will be included in this report.

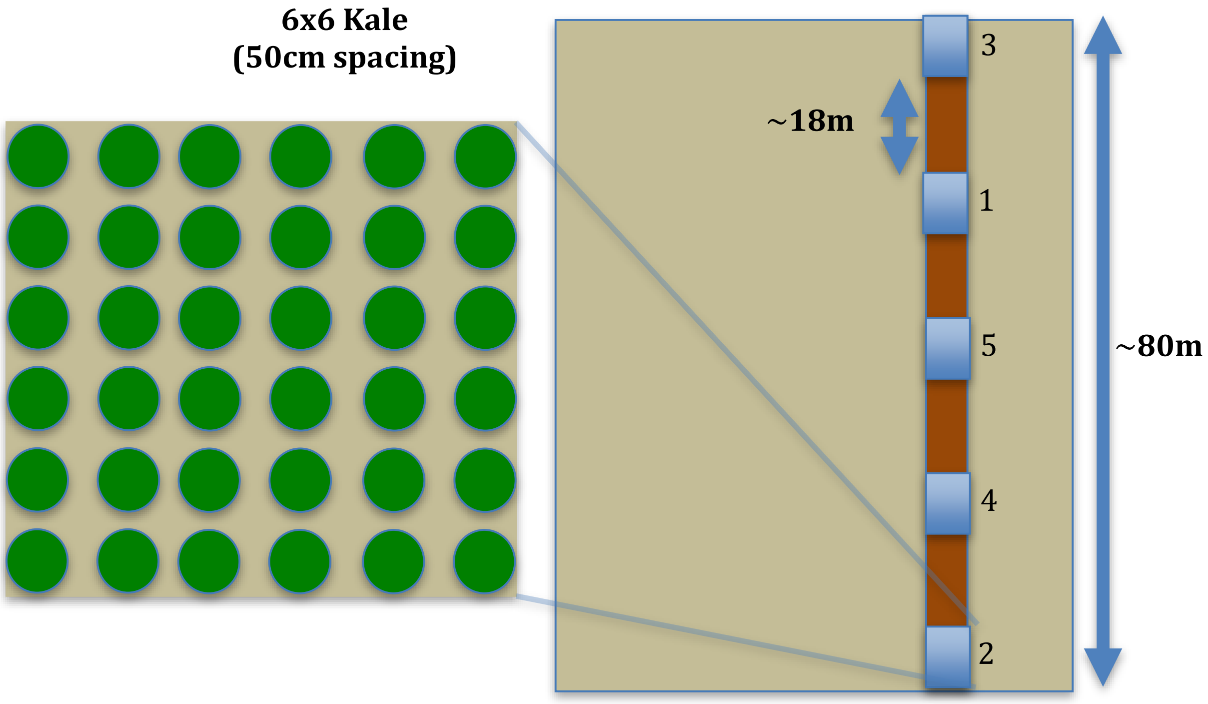

Figure 5. Schematic for plantings of kale plots at each of the five locations at Warwick Crop Centre, Wellesbourne. The numbers 1-5 represent the randomized positions of the sub-plots (planted May-September).

Results

Experiment 1. Monitoring of whitefly on a newly-planted vulnerable field crop (Brussels sprout and kale) throughout season.

Populations of whitefly remained very low for the first few months after transplanting, it was not until June that an increase in the numbers of eggs occurred, and soon after that all other life stages increased dramatically in numbers (Figures 6-9). This trend continued until September when numbers ceased to increase. All populations decreased from November until January when a large increase in egg numbers was observed. The corresponding increase in numbers of nymphs did not occur until March and finally, an increase in the numbers of pupae occurred in mid May. The last peak in numbers observed on these plots was the dramatic increase in adult numbers in early June, after which all life stages declined to zero in most cases.

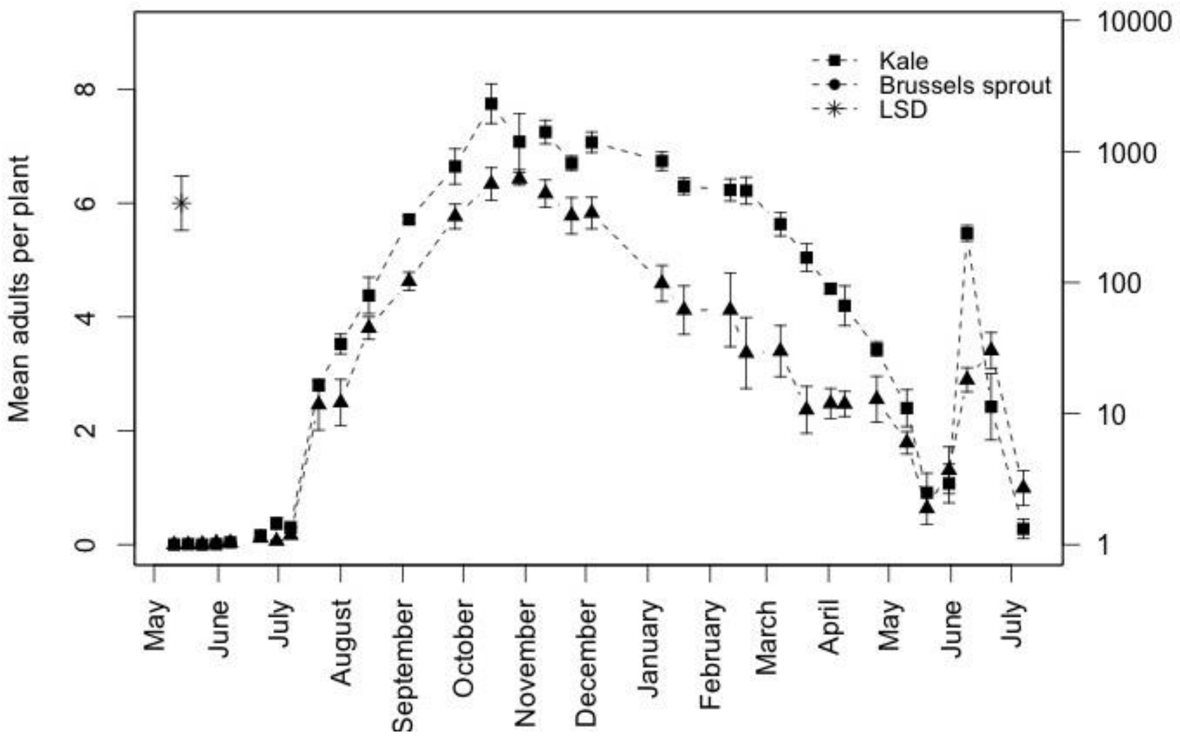

Figure 6. Mean (±SE) number of adults per plant on kale and Brussels sprout from May 2013 until July 2014 for the 5 study plots. The 5% LSD from ANOVA (P<0.01) is also shown. Left axis shows log+1 transformed data while the right axis shows untransformed data.

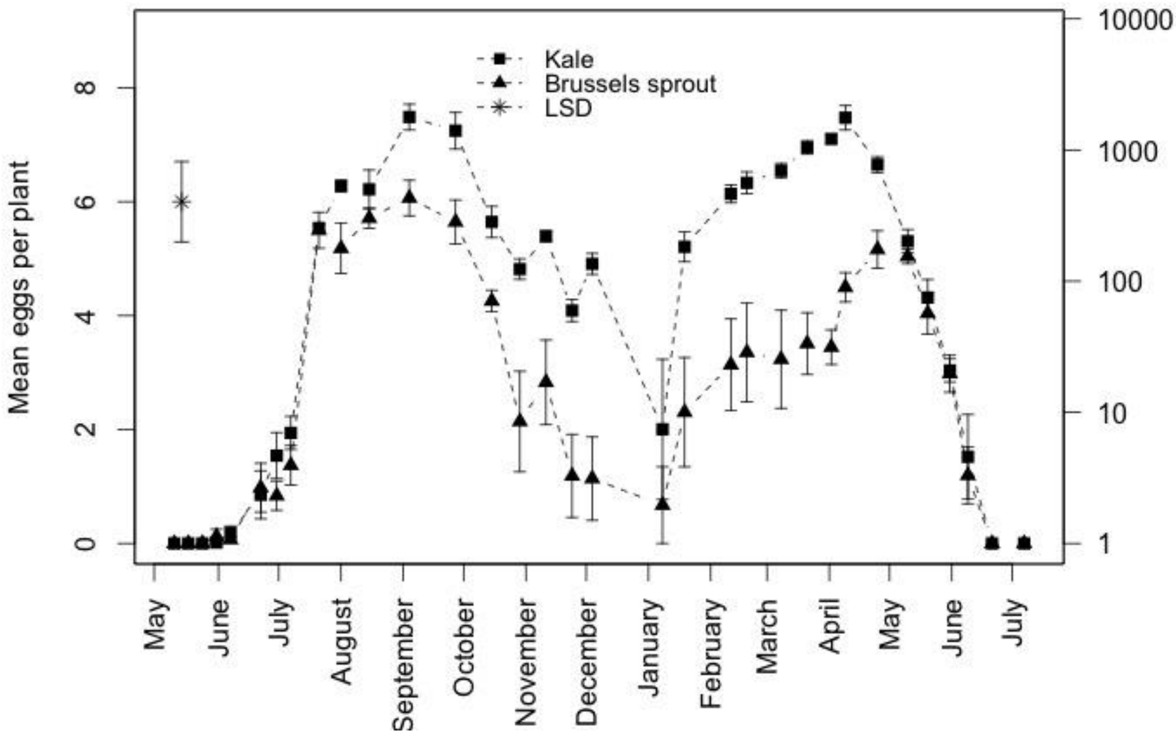

Figure 7. Mean (±SE) number of eggs per plant on kale and Brussels sprout from May 2013 until July 2014 for the 5 study plots. The 5% LSD from ANOVA (P<0.01) is also shown. Left axis shows log+1 transformed data while the right axis shows untransformed data.

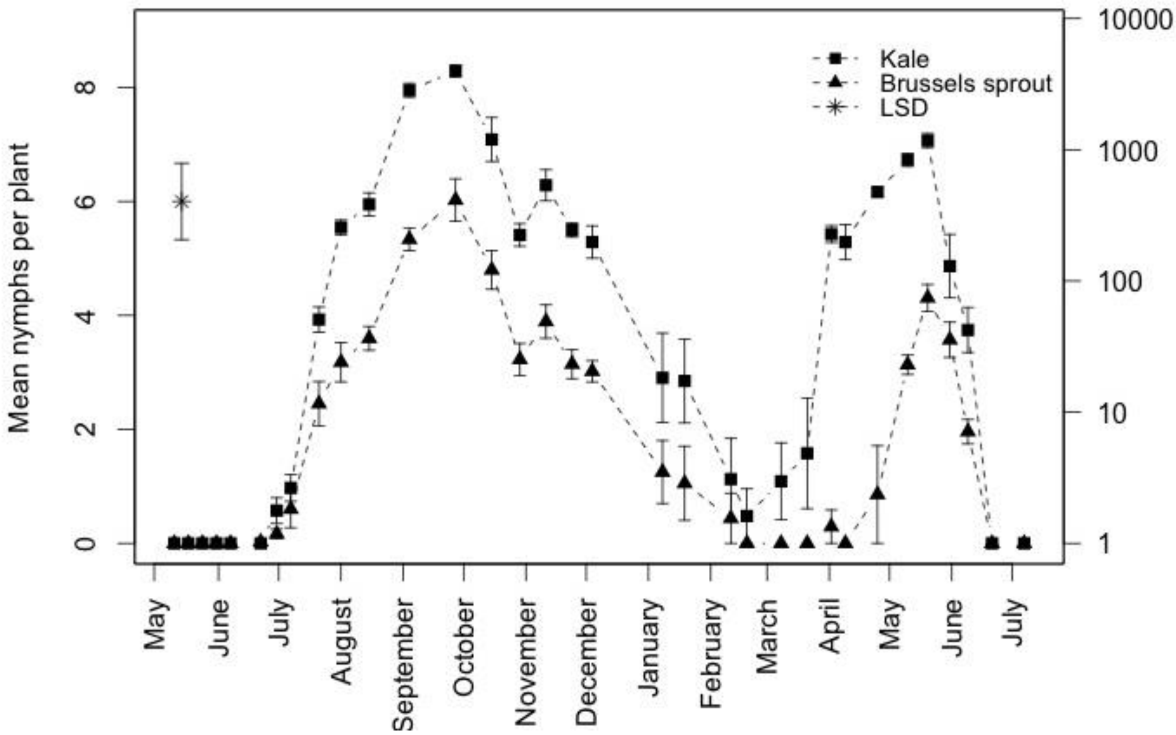

Figure 8. Mean (±SE) number of nymphs per plant on kale and Brussels sprout from May 2013 until July 2014 for the 5 study plots. The 5% LSD from ANOVA (P<0.01) is also shown. Left axis shows log+1 transformed data while the right axis shows untransformed data.

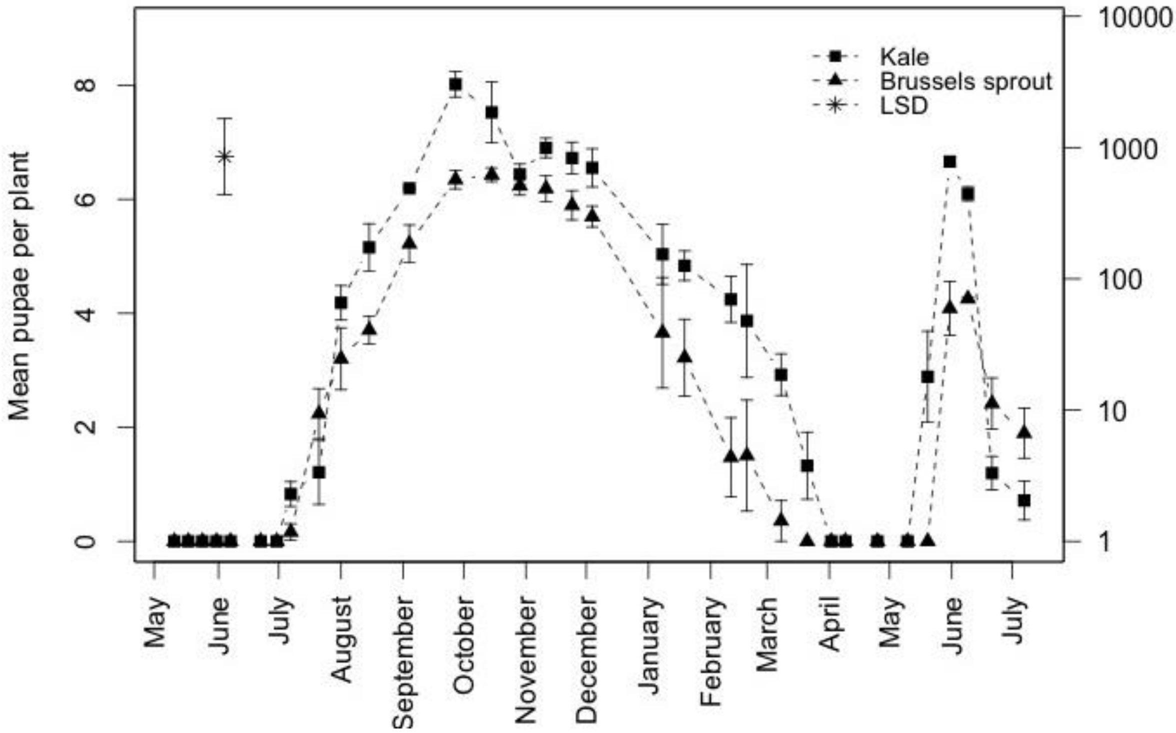

Figure 9. Mean (±SE) number of pupae per plant on kale and Brussels sprout from May 2013 until July 2014 for the 5 study plots. The 5% LSD from ANOVA (P<0.01) is also shown. Left axis shows log+1 transformed data, the right axis shows untransformed data.

Comparing cultivars.

Initial populations on Brussels sprout and kale did not differ significantly but subsequently, kale had consistently higher populations of all life stages; eggs (Figure 7), nymphs (Figure 8), pupae (Figure 9) and adults (Figure 6) after August.

Comparisons between plots.

One the whole, when considering each crop separately and comparing the numbers of each life stage, there were few differences between the plots.

Brussels sprout

The numbers of pupae found on the Brussels sprout plots were generally similar, with occasional statistically-significant differences between plots. For example, in August 2013, plot B had fewer pupae than the other plots, but this difference was no longer evident at the next sampling date (Figure 10). For adults, the only significantly different plot was plot C, which had a lower population in February 2014 (Figure 11)

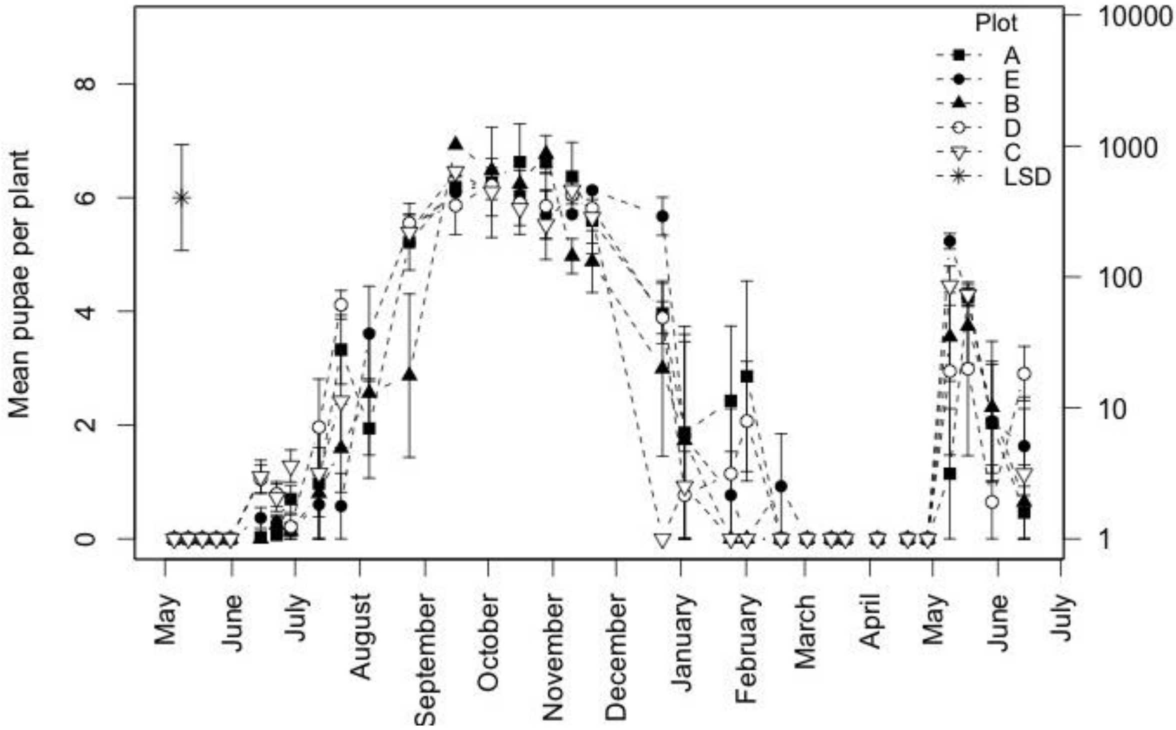

Figure 10. Mean (±SE) number of pupae per plant on Brussels sprout from May 2013 until July 2014 for the 5 study plots, A,B,C,D,E (see figure 3). The 5% LSD from ANOVA (P<0.01) is also shown. Left axis shows log+1 transformed data while the right axis shows untransformed data.

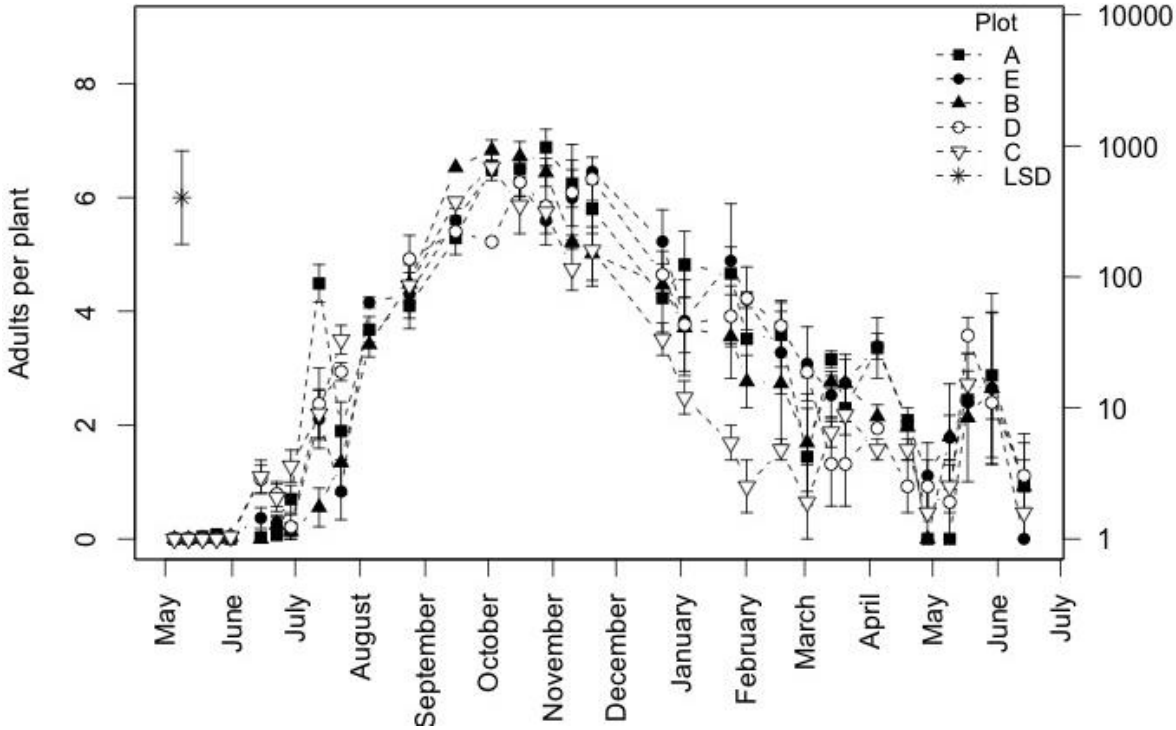

Figure 11. Mean (±SE) number of adults per plant on Brussels sprout from May 2013 until July 2014 for the 5 study plots, A,B,C,D,E (see figure 3). The 5% LSD from ANOVA (P<0.01) is also shown. Left axis shows log+1 transformed data while the right axis shows untransformed data.

Kale

There were some differences in the numbers of adults on the 5 plots of kale between May 2013 and July 2014 but these were not consistent. The most notable difference was between plots D and E in September 2013, but this difference had disappeared by the next sampling date (Figure 12).

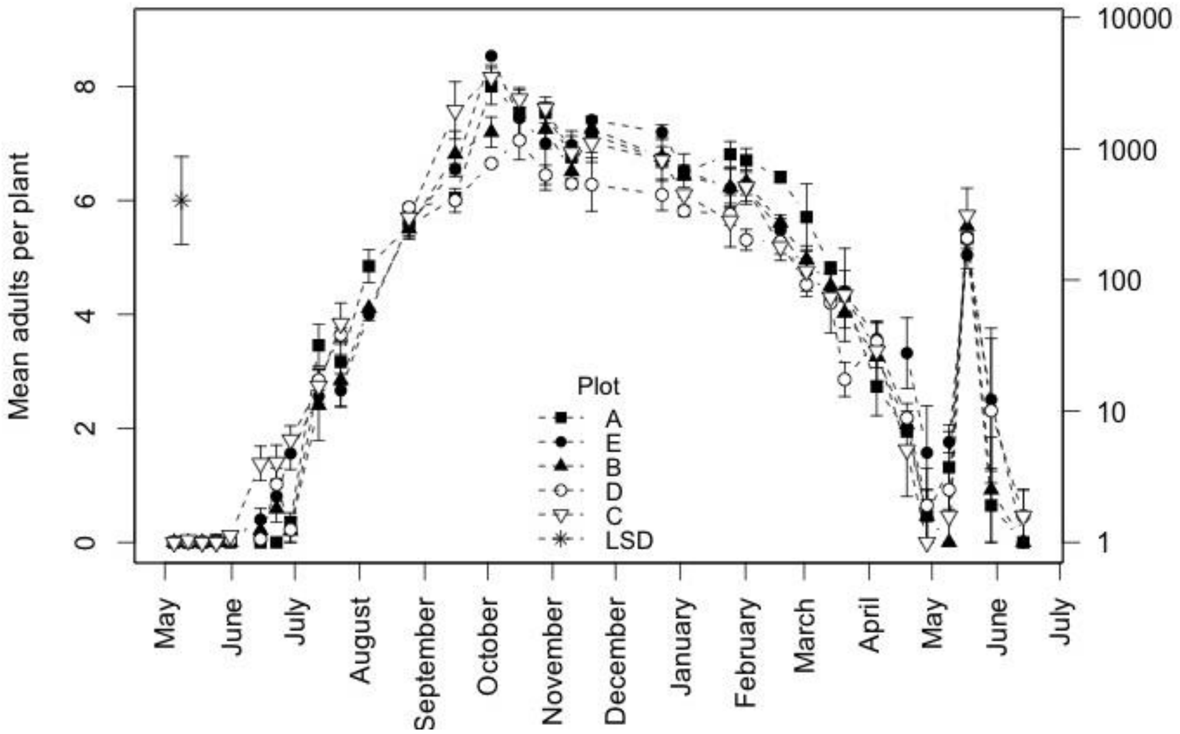

Figure 12. Mean (±SE) number of adults per plant on kale from May 2013 until July 2014 for the 5 study plots, A,B,C,D,E (see figure 3). The 5% LSD from ANOVA (P<0.01) is also shown. Left axis shows log+1 transformed data while the right axis shows untransformed data.

Egg numbers on the plots of kale were very similar for the majority of the sampling dates and there were generally no statistically-significant differences. The only significant differences between plots were over the winter period when eggs were still present on plots C and E but the other plots had none (Figure 13).

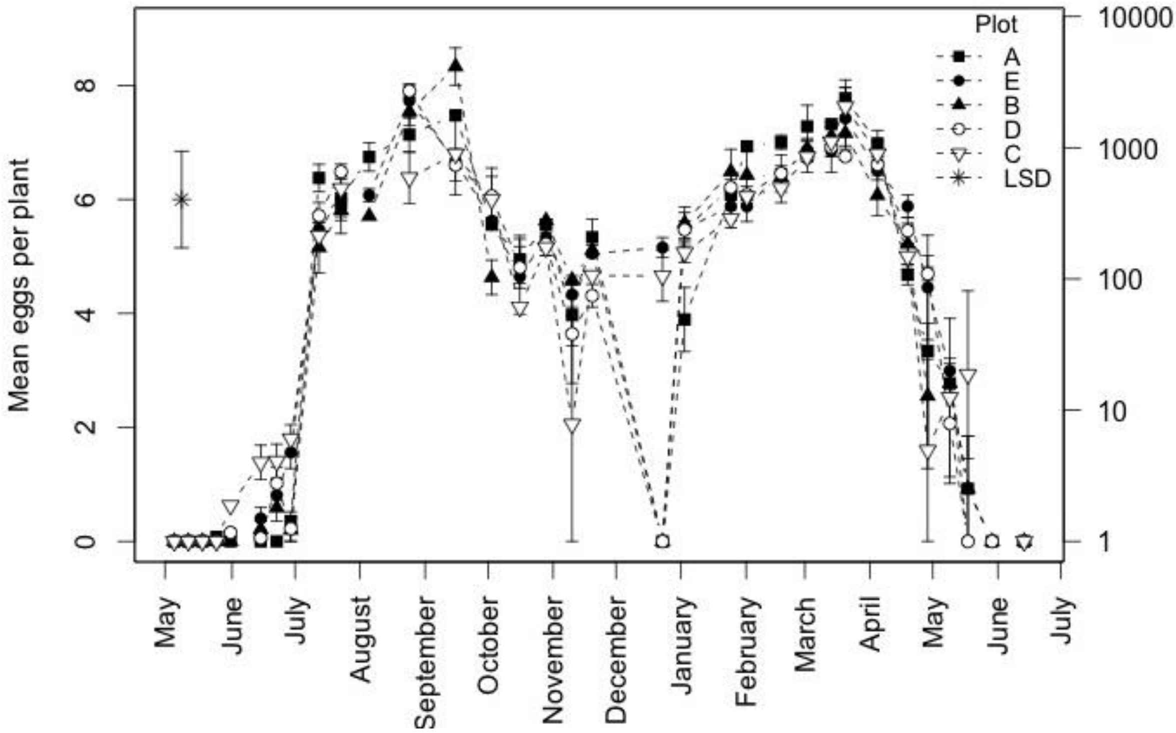

Figure 13. Mean (±SE) number of eggs per plant on kale from May 2013 until July 2014 for the 5 study plots, A,B,C,D,E (see figure 3). The 5% LSD from ANOVA (P<0.01) is also shown. Left axis shows log+1 transformed data while the right axis shows untransformed data.

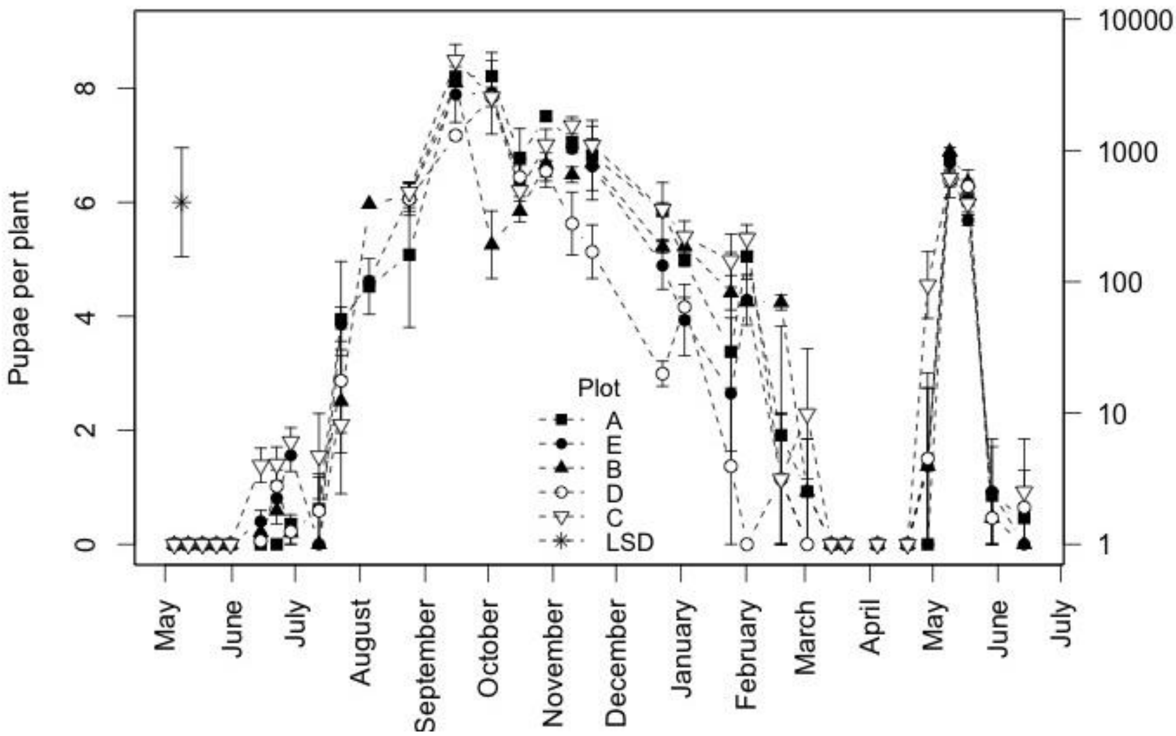

Figure 14. Mean (±SE) number of pupae per plant on kale from May 2013 until July 2014 for the 5 study plots, A,B,C,D,E (see figure 3). The 5% LSD from ANOVA (P<0.01) is also shown. Left axis shows log+1 transformed data while the right axis shows untransformed data.

On kale, the numbers of pupae per plant were very similar on all 5 plots between May 2013 and September 2013. It was not until the winter months that plot D had significantly lower numbers than 3 of the other plots (Figure 14). After March 2014 the numbers of pupae on each plot were again very similar, showing no significant differences.

To summarise, no plot had consistently lower numbers of any life stage than the others. Statistically-significant differences between plots in the numbers of whitefly were usually for only a short duration and most of the differences occurred over the winter months.

Experiment 2. Height at which adult whitefly disperse.

Yellow sticky traps caught significantly more whitefly than blue traps but this trend was only evident for heights up to 120cm above the ground (ANOVA, $P<0.01$), above these heights catches from blue and yellow traps did not differ. Yellow sticky traps at ground level caught significantly more adult whitefly than any other trap colour/height combination, catching on average ~50 individuals during the study period. The next highest catch rate was by yellow traps at 60cm and here the average catch rate fell to only ~5 individuals (Figure 15).

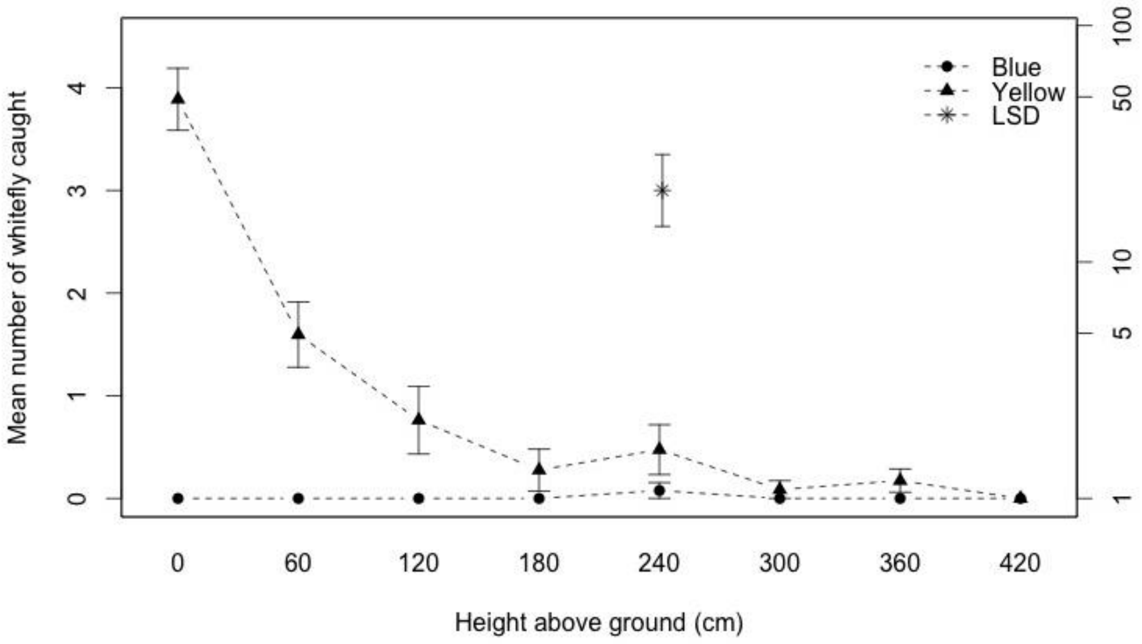

Figure 15. Mean (±SE) number of whitefly caught on yellow or blue sticky traps at different heights above ground. The 5% LSD from ANOVA is also shown ($P<0.01$). Left axis shows log+1 transformed data while the right axis shows untransformed data.

Experiment 3. Distribution of whitefly in a commercial crop of kale.

Sampling into the field showed that whitefly numbers near the edge were significantly different from those nearer the centre (ANOVA, P<0.01, Figure 16). Average numbers on plants at the edge were approximately 4 times greater than those at a distance of 115 plants into the field.

Comparing populations between each of the field edges (North, South, East and West) showed that numbers of adult whitefly differed between them (ANOVA, P<0.01). The most pronounced difference was between the northern and southern edges where there was an approximately a five-fold difference in numbers of adult whitefly (Figure 17).

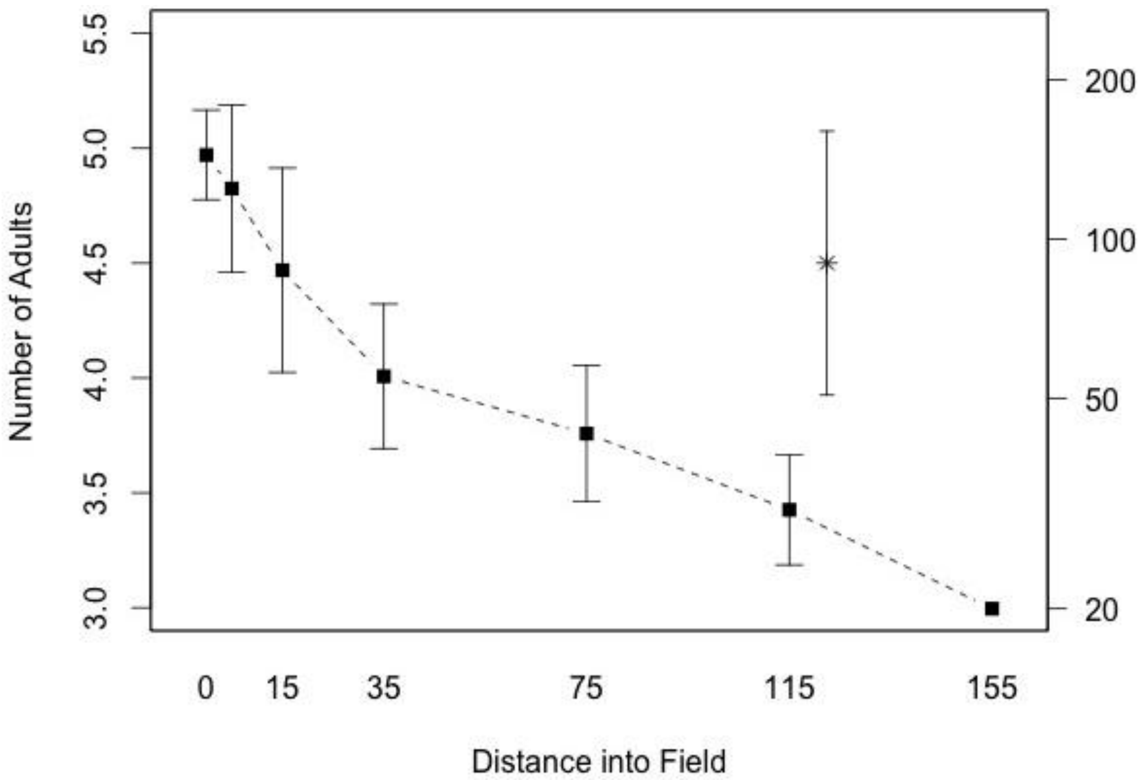

Figure 16 Mean (±SE) number of adult whitefly on 4 plants at different distances from each edge of a commercial crop of kale. The 5% LSD from ANOVA (P<0.01) is shown as an asterisk point. Left axis shows log+1 transformed data while the right axis shows untransformed data.

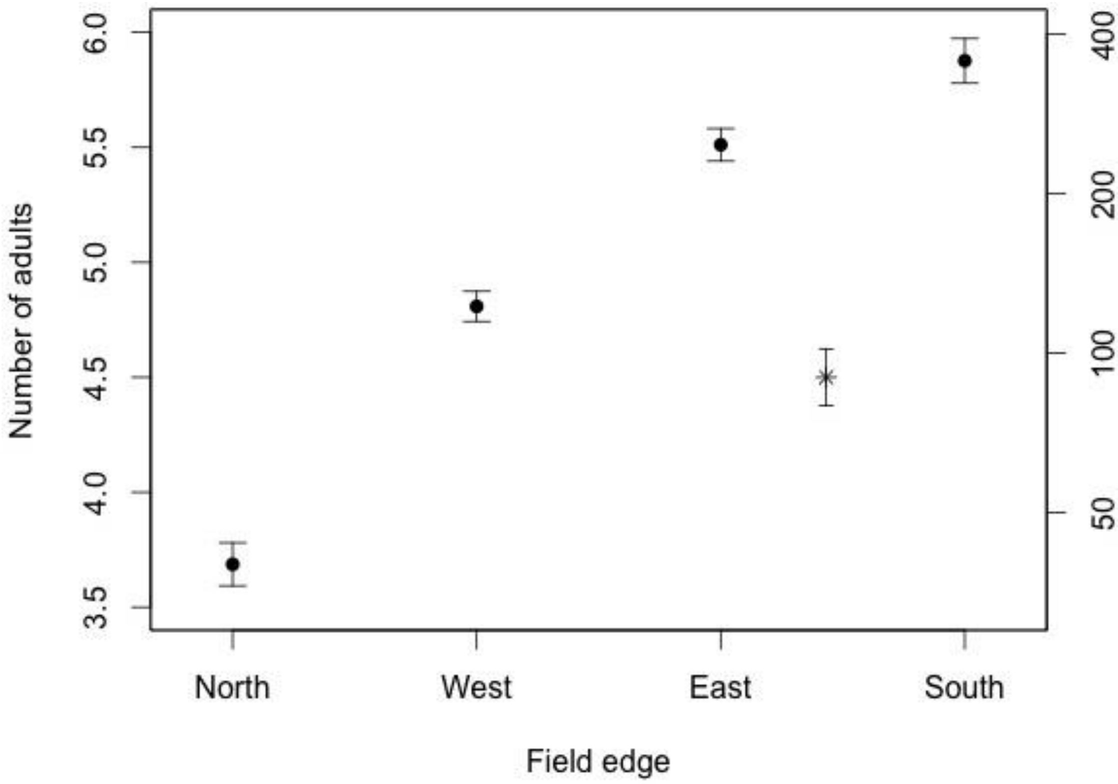

Figure 17. Mean (±SE) number of adult whitefly found on plants along each of the field edge (North, West, East and South). The 5% LSD from ANOVA (P<0.01) is also shown as an asterisk. Left axis shows log+1 transformed data while the right axis shows untransformed data.

Experiment 4. Distance transects of whitefly caught from known population source, sticky traps and trap plants.

Trap plants

The numbers of whitefly found on trap plants placed at various distances from a known population source were significantly different (ANOVA, P<0.01, Figure 18). There was a significant decrease in the number of adult whitefly found between 0 and 5 metres, but the numbers found on plants placed at distances of 5 and 15 metres did not differ. At 25 metres, the number of whitely was nearly 5 times less than that at 5 metres.

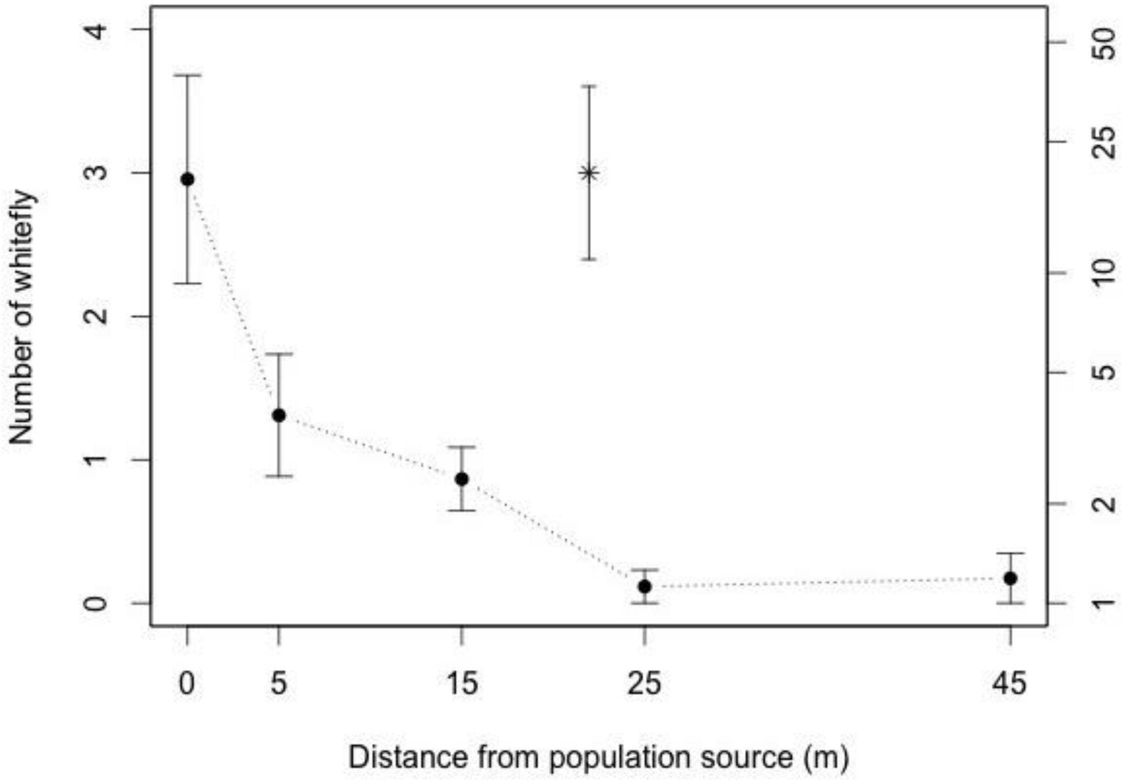

Figure 18. Mean (±SE) number of whitefly found on trap plants placed at different distances from a population source. The 5% LSD from ANOVA (P<0.01) is also shown as an asterisk. Left axis shows log+1 transformed data while the right axis shows untransformed data.

Yellow sticky traps

The number of whitefly caught on yellow sticky traps at distances of 0 and 5 metres from the population source differed significantly from each other and from all other traps within the transect. The numbers captured on traps located at the remaining distances of 10m, 20m and 30m caught very similar numbers of whitefly and did not catch significantly different numbers from each other (ANOVA, P<0.01, Figure 19).

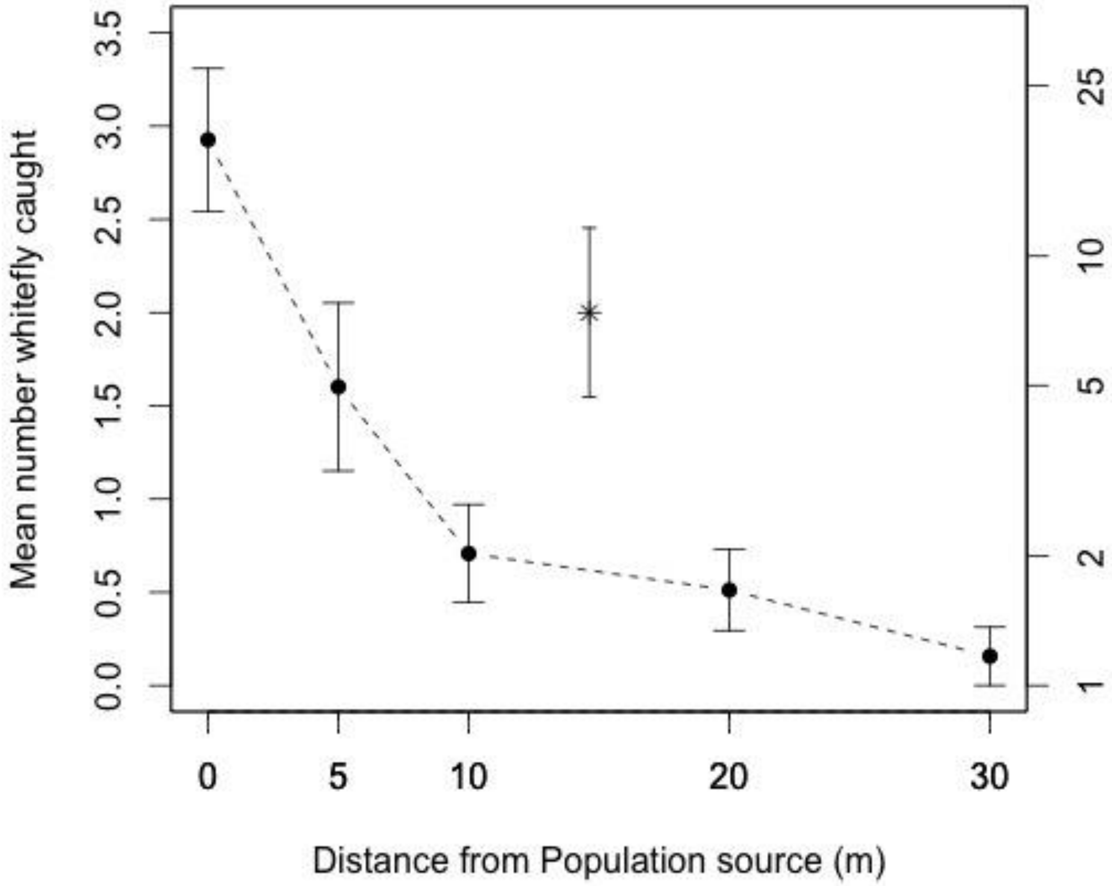

Figure 19. Mean (±SE) number of whitefly caught on yellow sticky traps placed on the ground at different distances from a population source. The 5% LSD from ANOVA (P<0.01) is also shown as an asterisk. Left axis shows log+1 transformed data while the right axis shows untransformed data.

Experiment 5. Monitoring of whitefly using yellow sticky traps placed on the ground.

An 'activity index' was calculated. This was defined as the log number of whitefly caught/mean adult whitefly per plant on plot. The 'activity index' varied during the year (August 2013 – July 2014). A slight, but statistically-significant peak occurred in October 2013. This was followed by a period where the activity index was at its lowest (January and February). The activity indices during the period April 2014 – July 2014 were significantly higher than at all other times, with a large, statistically-significant peak occurring in late May – June (Figure 20).

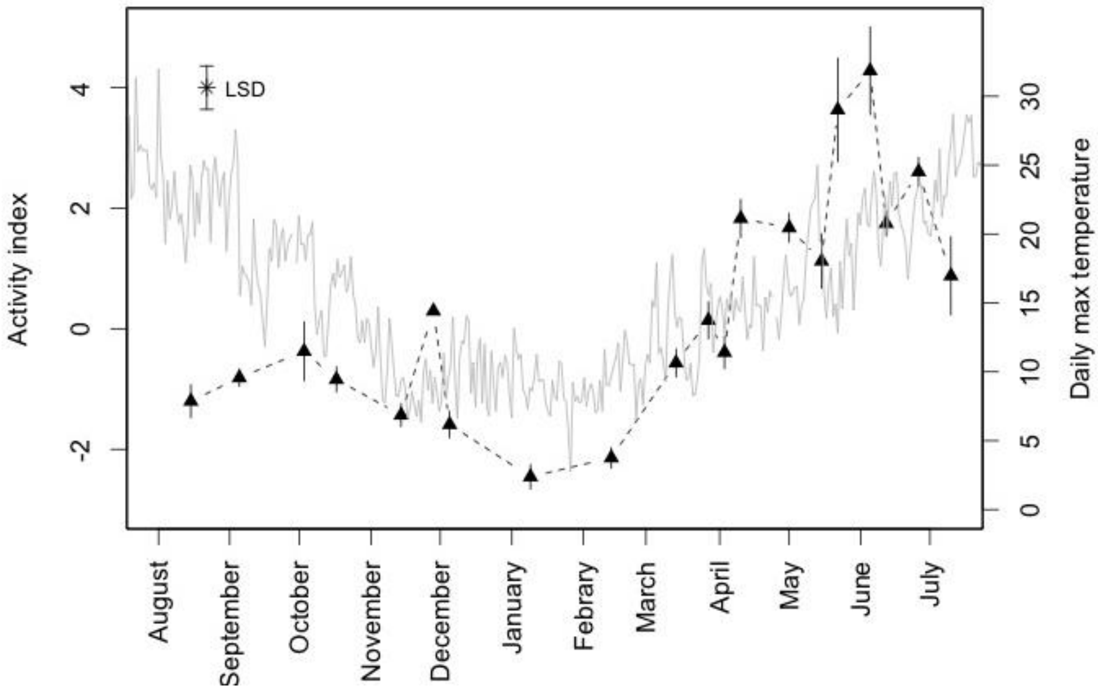

Figure 20. Mean (±SE) whitefly activity index [logged (Number whitefly caught on traps/numbers per plant in plot)] throughout a year (August 2013 – July 2014). The 5% LSD from ANOVA (P<0.01) is also shown. The grey line represents daily maximum air temperatures (°C) for Wellesbourne.

Throughout most of the year (August 2013 – July 2014) the catch rates for sticky traps to the North, South, East or West of the plots did not differ significantly from one another. There was a long period from December 2013 until March 2014 when catch rates were significantly higher on the traps to the North or East (ANOVA, P<0.01, Figure 21) than those to the South and West.

There was a significant relationship between the log sticky trap catch and the log mean adult population (log-log linear model y=0.9173x, P<0.01, R^2=0.88, y-intercept constrained to 0, Figure 22).

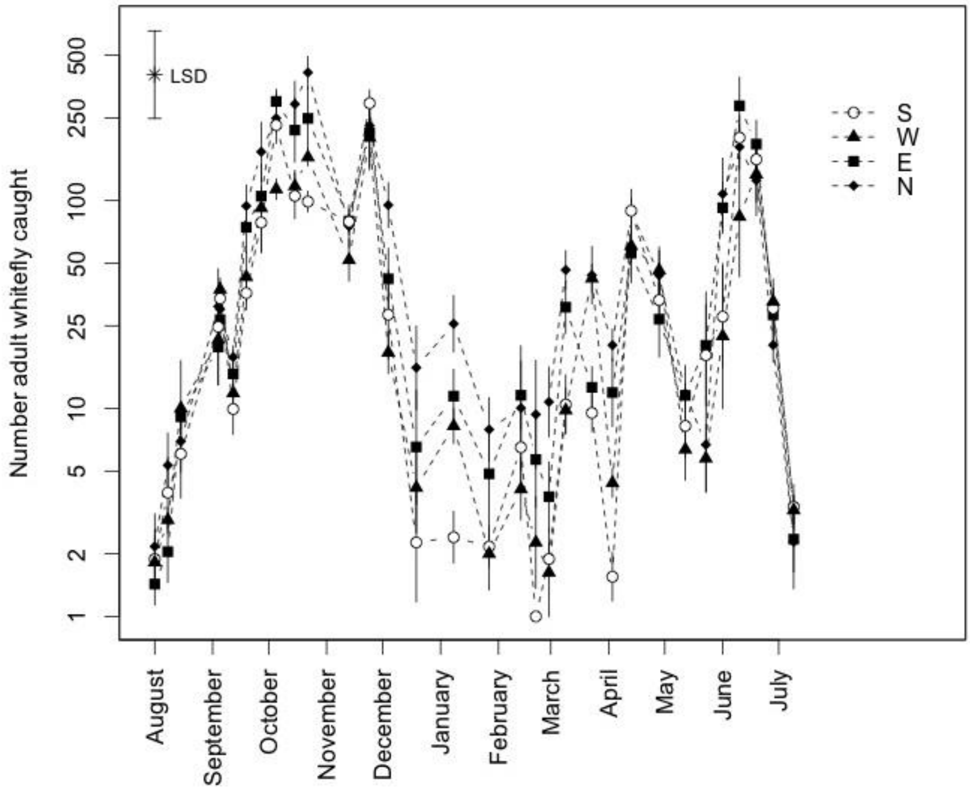

Figure 21. Number of whitefly caught on traps placed in different directions from plots (N-North, S-South, E-East, W-West) The 5% LSD from ANOVA (P<0.01) is also shown

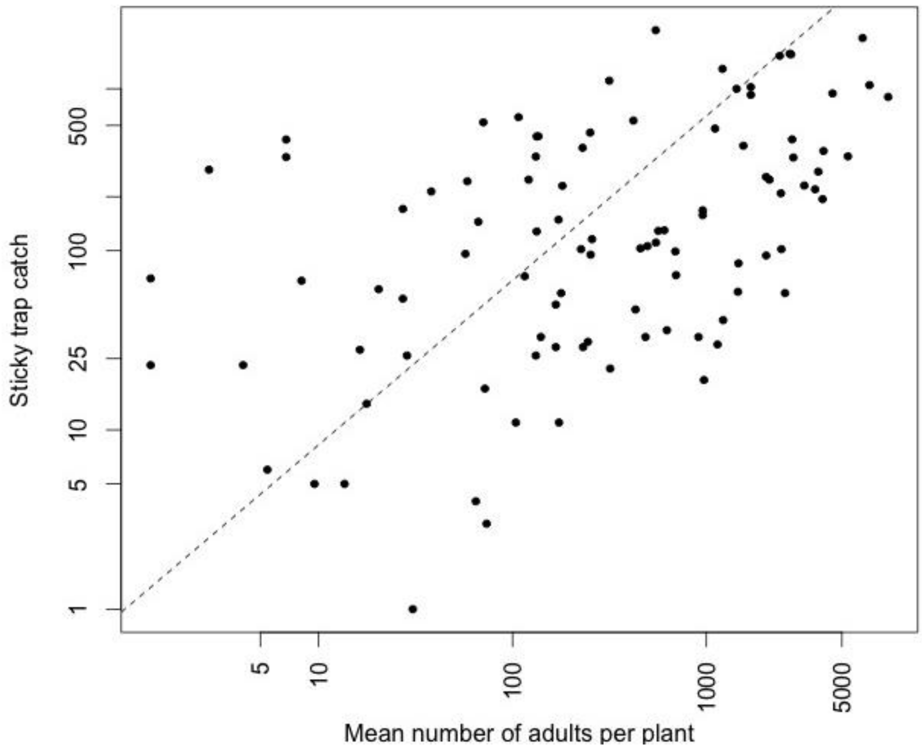

Figure 22. Mean number of whitefly caught on sticky traps versus the mean number of adults per plant. A fitted logged linear model (y=0.9173x) is also shown as a dotted line (P<0.01).

Experiment 6. **Monitoring of immigration and establishment of whitefly populations on spatially and temporally separated plantings of kale.**

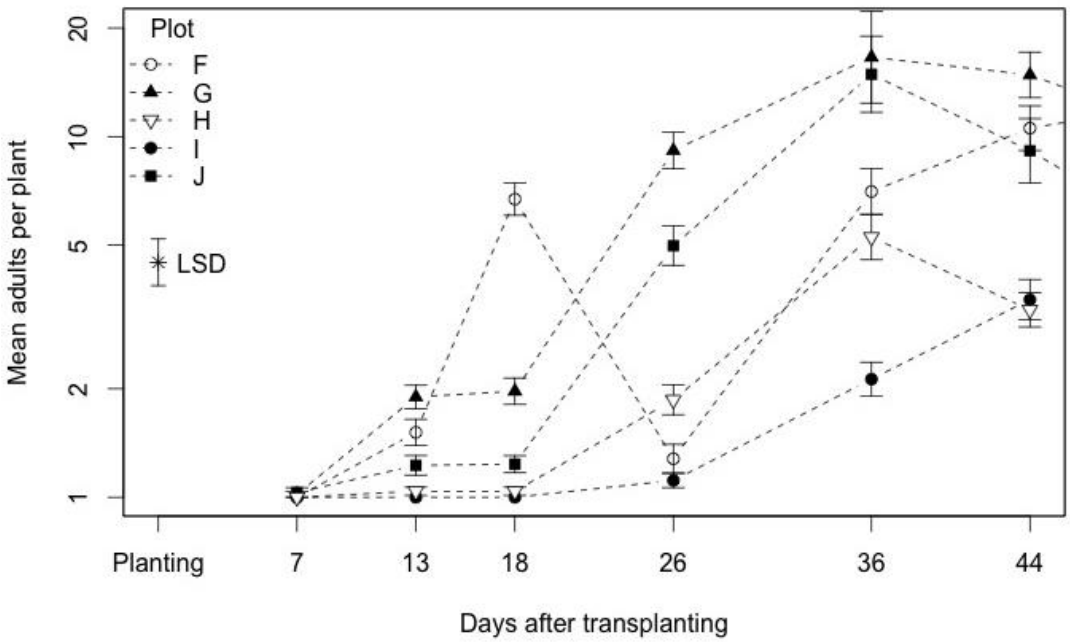

Figure 23. Mean (±SE) number adults per plant for the five study plot plantings made in May 2014. The 5% LSD from ANOVA (P<0.01) is also shown

The numbers of whitefly found on the kale plants transplanted in May 2014 differed between plots from as early as day 13 (ANOVA, P<0.01, Figure 23). Plots H and I had the lowest numbers and were only significantly different from each other on days 26 and 36. Plot G had the highest number of whitefly on most occasions and this was often significantly higher than all other plots. A decline in the number of adults was observed in plot F on day 26 due to a huge loss of foliage as a result of herbivory. All plants in this plot were replaced on day 30.

There was a statistically-significant relationship between the adult population on the newly-transplanted plot at day 13 and the distance of the plot from a highly-infested plot planted in 2013 (Figure 24). Doubling the distance from 50m to 100m led to an approximate reduction in the adult population of 75%.

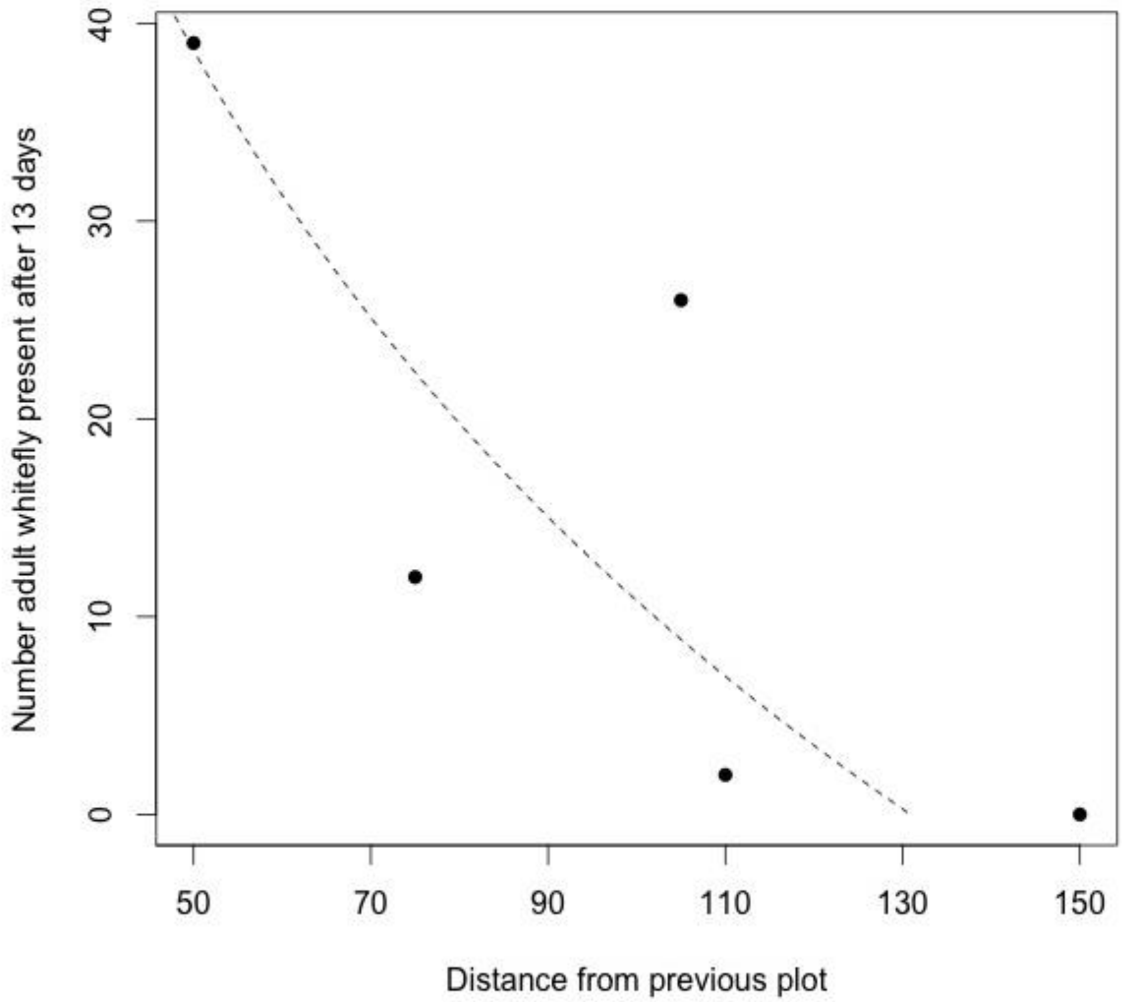

Figure 24. Number of whitefly present within the newly-transplanted plots, 13 days after transplanting versus the distance of each plot from the nearest plot planted in 2013. The dashed line shows a statistically-significant GLM (Population after 13 days ~Distance, family= poisson, $y=-0.02491x+4.87437$, $P<0.01$)**.**

Discussion

Experiment 1. Monitoring whitefly on newly-planted vulnerable field crops (Brussels sprout and kale) throughout season.

Although it was a very small number (0.04 adults per plant 36 days after planting), the first adult whitefly arrived on the plots in mid-May 2013. This was before the first generation emerged on an overwintering crop in the same location; on 28th June. (Year 1 report, Experiment 1.5 (Collins, 2013)). This provides evidence that the first females to colonise the plots were overwintering females.

The sizes of the initial adult populations were very similar between crops (kale and Brussels sprout), suggesting no difference in attractiveness of host to the immigrating whitefly. The number of eggs remained similar on both crops, suggesting a similarity in host quality for determining fecundity of arriving adult females. This observation supports the information collected in the laboratory in the first year of the project (Year 1 report, Experiment 1.4 (Collins, 2013)) where the number of eggs laid by females on kale and Brussels sprout plants did not differ significantly. Differences between the two crops became apparent after August, by when an entire generation would have developed on the crop. It could be that the emergent females from the two crops are of different quality and it would be interesting to test this in a laboratory studies in the future. Differences were most pronounced over winter, suggesting that overwintering females may prefer kale to Brussels sprout plants. It could be that the structure of kale plants provides more protection to the whitefly, causing them to be less easily disturbed by rain or wind, for example. It would be interesting to test whether females, especially those overwintering, are less likely to be dislodged by rain on kale when compared with Brussels sprout plants.

In general there was little difference in the number of all life stages of cabbage whitefly between the five plots. A lower number of adults was evident in plot C on the Brussels sprout plants. This plot was particularly damaged by caterpillars that significantly reduced the overall leaf area of the plants. This is likely to have had an impact on the availability of locations for whitefly to populate. Brussels sprout plants in general were more damaged as a result of herbivory by caterpillars and this reduced leaf surface area may have partly contributed to lower numbers of whitefly seen on the Brussels sprouts. This cannot be the complete cause of the difference in numbers as whitefly never populated 100% of the leaves of the Brussels sprout plants and therefore leaf surface area was very unlikely to be the limiting factor.

Egg laying by overwintering females first occurred in January 2014. This was a month earlier than in 2013, when the first eggs were not seen until mid February (Year 1 report, Experiment 1.5, (Collins, 2013)). The winter of 2014 was considerably warmer than 2013 with mean January maximum temperatures of 6.8°C in 2014, compared with 4.6°C for 2013. This may be the reason for earlier egg laying. Laboratory studies will be conducted to try and ascertain the low temperature threshold for egg laying, which, when determined, can be used to see if the date of 'first egg lay' can be predicted. Although the first eggs were laid in January, nymphs were not observed until the end of March; so either the subsequent low temperatures reduced the development of eggs to such a level that it took nearly 2 months for them to hatch, or the first cohort of eggs died as a consequence of the low temperatures. Currently laboratory work is being conducted to ascertain whether egg mortality occurs after prolonged exposure at 0^{o}C. It would also be interesting to see how long eggs can lie 'dormant' at temperatures below their lower development threshold (<10°C, Iheagwam, 1978), as this may be more relevant to field conditions.

The numbers of all life stages began to decrease dramatically from June 2014 as the plants were undergoing senescence following flowering and seed production. The declining leaf area provided little or no habitat for any life stage and this is likely to have been the cause for decline. Studies on the rate of development of the different life stages at different temperatures are on-going and, once they have been completed, a simple day degree model will be created. Data from this experiment will be used to validate the model.

Experiment 2. Height at which adult whitefly disperse.

The results from this experiment support those from Experiment 1 which was described in the first year report (Collins, 2013). Yellow sticky traps placed at ground level are the most successful at capturing adult cabbage whitefly. Due to the interaction of colour and height seen in the same experiment in Year 1 (Collins, 2013) it was hypothesised that blue might become a more preferred colour at greater heights above the ground. This was not the case. However, there was a slight increase in the numbers of whitefly captured on blue sticky traps suspended at a height of 240cm. Further replication of this experiment is required to give the statistical analysis enough power to distinguish between these slight differences in catch rates.

Data shown in this report only represent the behaviour of reproductively active whitefly. However, previous studies have suggested that female cabbage whitefly entering reproductive diapause show stronger migratory behaviours (Iheagwam, 1977b). The current study will be replicated at times when females in diapause are present; during September and October, for example. As migration in insects is often distinguished by a shift from preference for host vegetative cues, i.e. yellow light, to that of sky light, i.e. Ultraviolet-Blue (Dingle, 1996), a change in colour preference to blue or greater height of flight would support the hypothesis that long range migration is completed by females who have just entered diapause.

These results again show the importance of trap height in terms of the efficacy of yellow sticky traps for monitoring the cabbage whitefly. A difference of 60cm can reduce catch rates of whitefly by ten-fold. Any sticky traps used for monitoring cabbage whitefly are likely be much more effective at ground level.

Experiment 3. Distribution of whitefly in a commercial crop of kale.

A clear 'edge effect' has been shown in this experiment. The numbers of whitefly in a field were significantly higher on plants on the edge of the crop than on those towards the centre. This study showed that 4 times as many adult whitefly were found at the edge of a field when compared to a distance of 45m into the crop (75 plants into field at 60cm spacing). The numbers of whitefly on each field edge were very different. The southern edge had a mean number of whitefly which was ten times that of the northern edge. This type of information is very important for growers who are surveying fields to ascertain whether they need to control whitefly. If counts are always made from the same edge of the field then growers are unlikely to get a true representation of the field as a whole. Further research will be needed on multiple fields and at multiple times of year, to see if this can be generalised to different field situations. The aspect of the field edge may also be important, and further sampling will be undertaken to investigate whether the southern edge is consistently more heavily-infested or whether this was an effect of locality. The southern edge of the field sampled was adjacent to a residential area, with some wooded areas while the northern edge was next to a dyke leading onto a potato field. The habitats that are near to field edges will also be investigated to see if this has any effect on whitefly numbers in the adjacent crop. Over the year the average wind direction in Spalding, Lincolnshire is East-South-East. Whitefly that may have been moved extra distances with the wind would be likely to land in at the eastern and southern field edges. This conforms to the two sides with the highest populations in the field. Further studies will need to be conducted to see if this pattern occurs in other fields.

Experiment 4. Estimating the abundance of adult whitefly at different distances from a field infestation.

As expected, counts of whitefly closest to areas of infestation were significantly higher than those at greater distances. What is particularly interesting, however, is the fact that for the trap plant study, the catches at distances of 25 and 45m did not differ from each other. A 20m increase in distance of the trap from the source of whitefly had no impact on numbers caught. Determining the distance when a population source no longer impacts catch rates would be important to indicating a potential minimum distance at which vulnerable crops should be planted from currently infested crops to reduce the impact of immigration from these sources.

Experiment 5. Monitoring whitefly activity using yellow sticky traps on the ground.

For the majority of the year, the direction in which the sticky trap was placed (i.e. northern, southern, eastern and western sides of the plot) had no effect on the catch rate. However, during December and January more whiteflies were captured on the traps placed to the North and the East. This also corresponds to the period when the whitefly had the lowest activity index. It is likely that trap captures at this time of year are whitefly that have been dislodged from their host rather than actively leaving the crop. The wind direction at Wellesbourne is predominantly south-westerly, this would cause any whitefly that are dislodged to be blown towards the northern and eastern traps more often.

The graph showing the activity index of whitefly throughout the year (Figure 20) indicates some interesting differences. First of all, there was an increase in activity in October. Female whitefly emerging at this time are known to have entered reproductive diapause. Previous work has shown that females within reproductive diapause show stronger flight behaviour (Iheagwam, 1977b) and the increase in the activity index supports this. As expected, the coldest months of December and January were also those showing the lowest levels of activity. The peak in activity in November is difficult to explain. There was also a slight increase in maximum temperature around the same time, which may have increased activity during this sampling period. A similar study is being conducted on the plots used in Experiment 6 and it will be interesting to see whether a similar peak occurs in 2014.

Activity of overwintering females increased through March until May. The higher activity of females early in the season shows the potential at this time for immigration into crops from overwintering sources. A sharp increase in activity in May-June occurred at a time when the first male whitefly were noticed on the traps, which signified the emergence of the first generation of adults (progeny of the overwintering females). The considerable increase in activity at the time that the first generation adults emerged supports the notion that first generation adults are likely to move onto new hosts and it signals a time when new crops are likely to be colonised. Although there was a significant relationship between the numbers of whitefly caught on sticky traps and the numbers of whitefly per plant (both in log numbers), sticky trap catches are unlikely to provide reliable estimates of the size of infestations in crops, particularly because they give no indication of the numbers of nymphs within the crop. Physically checking leaves is the only method that could provide information on the numbers of nypmhs present within the crop, which informs the grower of both current adult populations and potential future populations.

Experiment 6. Monitoring immigration and establishment of whitefly populations on spatially- and temporally- separated plantings of kale.

Whitefly numbers increased quickly in this study; the mean number of whitefly per plant in Plot F exceeding 10 after only 36 days. In contrast, this did not happen in 2013 (in Experiment 1) until after 2 months (Figure 6). One factor that may have contributed to this difference was the larger overwintering whitefly population at Wellesbourne in 2013-14 than in 2012-13. The plots from the 2013 study were still present in early 2014 and were likely to have been the main source of immigration into the 2014 plots. The statistically-significant model relating the numbers of whitefly on each new plot after 13 days to its distance from the nearest source of whitefly (plots planted in 2013) supports this (Figure 24). The closer a 2014 plot was to a plot planted in the previous year, the higher rates of immigration it received. Doubling the distance from the source from 50m to 100m seemed to cause a 75% reduction in the numbers of whitefly (after 13 days). This suggests, a very reasonable conclusion, that the rate of colonisation by whitefly onto new crops is highly influenced by the distance of the new crop from sources of overwintering females. Such locations are likely to be overwintered brassica crops such as kale, cauliflower and oilseed rape.

This experiment is on-going and, for example, relationships between the colonisation rates and the time of planting will also be investigated. Inter-plot differences will also be monitored to ascertain whether the differences observed between the May plantings continue throughout the season.

Conclusions

- Kale and Brussels sprout plants appear to have the same level of 'attractiveness' to colonising cabbage whitefly.
- Egg laying began a month earlier in 2014 than in 2013. Mean January maximum daily temperatures were 2°C warmer in 2014 than 2013 and this might have been the reason for the earlier egg laying.
- Field edges supported, on average, 4 times as many whitefly as areas towards the centre of the field. Populations of whitefly at each field edge also differed significantly from each other.
- Whitefly are most effectively caught on yellow sticky traps at ground level. Increasing the height to 60cm led to, on average, a ten-fold reduction in catch rate.
- Activity of overwintering female whitefly increased during February-March showing potential for these females to move into new crops.
- Peak activity of adult whitefly was in May-June when the first generation of adults where emerging. It is likely to be the time of greatest colonisation of new crops.
- The rate of colonisation of new crops by whitefly is highly influenced by the distance of the new crop from sources of overwintering females. An increase in distance from 50m to 100m led to a 75% reduction in immigration rate.
- Sticky traps are unlikely to provide reliable estimates of whitefly populations on field crops, as they do not provide information on juvenile stages, and other factors such as weather conditions and the reproductive condition of females are likely to influence catch rate.

Knowledge and Technology Transfer

- October, 2013. Attended Brassica growers association R&D meeting, Lincolnshire
- November 2013 Presented at Brassica Technical Seminar, Lincolnshire
- January, 2014 Attended Brassicas growers association Conference, Lincolnshire
- March 2014 Presented poster and abstract in University of Warwick student symposium
- September 2014 Presentation given as part of Tomato growers association student conference. Poster also presented, Warwickshire.

References

Adams, A.J. (1985a) The photoperiodic induction of ovarian diapause in the cabbage whitefly, *Aleyrodes proletella* (Homoptera: Aleyrodidae). Journal of Insect Physiology. 31:693-700.

Adams, A.J. (1985b) The critical field photoperiod inducing ovarian diapause in the cabbage whitefly, *Aleyrodes proletella.* (Homptera: Aleyrodidae). Physiological Entomology. 10:243-249.

Al-Houty, W. (1979) Some ecological studies on the cabbage whitefly, *Aleyrodes brassicae* (Walker) Hemiptera-Homoptera-Aleyrodidiae. Ph.D thesis, Bath University.

Butler, C.G. (1938a) On the ecology of *Aleurodes brassicae* Walk. (Hemiptera) Transactions of the Royal Entomological Society of London. 87:291-311.

Butler, C.G. (1938b) A Further contribution to the ecology of *Aleurodes brassicae.* Walk. (Hemiptera). Proceedings of the Royal Entomological Society of London. 13:10-12. 161-172.

Collins, S. (2013) Biology of the cabbage whitefly (*Aleyrodes proletella*). Year 1 report. AHDB Horticulture.

DEFRA. (2012) Basic horticultural statistics, 2012. Department for Evironment, Food and Rural Affairs.

El-Khidir, E. (1963) Ecological studies on *Aleyrodes brassicae* with special reference to dispersal. Ph.d. thesis. University of London.

Garthwaite, D.G., Barker, I., Parrish, G., Smith, L., Hudson, S. & Pietravalle, S. (2013) Pesticide survey report, 257. Outdoor vegetable crops in the United Kingdom, 2013. Department for Environment, Food and Rural Affairs

Gerling, D. (1990) Whiteflies: their Bionomic, Pest Status and Management Intercept, Andover,Hants,UK.

Iheagwam, E.U. (1977a) Photoperiodism in the cabbage whitefly, *Aleyrodes brassicae*. Physiological Entomology. 2:179-184.

Iheagwam, E.U. (1977b) Comparative flight performance of the seasonal morphs of the cabbage whitefly, *Aleyrodes brassicae* (Wlk), in the laboratory. Ecological Entomology. 2:267-271.

Iheagwan, E.U. (1978) Effects of temperature on the development of the immature stage of the cabbage whitefly, *Aleyrodes proletella* (Homoptera: Aleyrodidae). Entomologia Experimentalis et Applicata*.* 23:91-95.

Martin, J.H., Mound, L.A. 2007. An annotated check list of the world's whiteflies (Insecta: Hemiptera: Aleyrodidae). Zootaxa, 1492: 1-84.

Nebreda, M., Nombela, G. & Muniz, M. (2005) Comparative Host Suitability of Some *Brasscia* Cultivars for the Whitefly, *Aleyrodes proletella* (Homoptera: Aleyroidae). Entomologia experimentalis et applicata. 23:91-95.

Richter, E. (2010). Population dynamics and chemical control of Aleyrodes proletella in vegetable brassica crops. In *57. Deutsche Pflanzenschutztagung, Berlin, Germany, 6-9 September, 2010.* (No. 428). Julius Kühn Institut, Bundesforschungsinstitut für Kulturpflanzen.

Springate, S. & Colvin, J. (2011) Pyrethroid insecticide resistance in British populations of the cabbage whitefly, *Aleyrodes proletella.* Pest Management Science. 68:260-267.

