Project title:	Managing ornamental plants sustainably (MOPS)
Project number:	CP 124
Work package title:	Efficacy of plant protection products against sucking insects – glasshouse whitefly / protected ornamental
Work package leader:	Dr Dave Chandler, Warwick Crop Centre
Report:	Annual report, December 2014
Previous report:	None
Key staff:	Dr Dave Chandler, Warwick Crop Centre Gill Prince, Warwick Crop Centre Andrew Mead, Rothamsted Research (statistical advice)
Location of work:	Warwick Crop Centre, Wellesbourne, Warwick, CV35 9EF
Date work commenced:	20th May 2014
Date work completed (or expected completion date):	19th September 2014

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.

G Prince
Research Fellow
Warwick Crop Centre

Signature Date 29/01/2015

Report authorised by:

Dr D Chandler
Senior Research Fellow
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Growers Summary

Headline

Six products were identified that caused significant reductions in populations of glasshouse whitefly feeding on verbena plants. The products appear to have worked mainly by causing death of whitefly during the nymphal and / or pupal stages, resulting in a reduction in the numbers of adult whitefly emerging from pupae.

Background and expected deliverables

Glasshouse whitefly (*Trialeurodes vaporariorum)* is one of the most common pests of ornamentals. Infested plants become contaminated with sticky honeydew excreted by whiteflies and this allows the growth of sooty moulds. In severe infestations, leaf yellowing and plant stunting occurs. The presence of whiteflies and damage symptoms can cause ornamental plants to be unmarketable. The glasshouse whitefly has developed resistance to pyrethroids such as deltamethrin (e.g. Decis) and pyrethrum (e.g. Spruzit) and there has been one recorded incidence of resistance to neonicotinoid insecticides such as imidacloprid (e.g. Intercept 70 WG) in the UK.

The purpose of Objective 2 was to test the efficacy of plant protection products against sucking insects. In particular, Objective 2.1 was to test the efficacy of new conventional chemical and biopesticide products against glasshouse whitefly *Trialeurodes vaporariorum* on a selected susceptible protected ornamental species.

Summary of the work and main conclusions

Seven plant protection products (Table 1) were tested against glasshouse whitefly (*Trialeurodes vaporariorum)* on Verbena plants maintained under glasshouse conditions between June and September 2014 at Warwick Crop Centre, Wellesbourne, UK. The glasshouse compartment was fitted with insect-proof screens in order to minimise the risk of plants becoming infested with other insect pests. Temperature within the compartment was regulated by venting the compartment at 15^{O}C and using additional heating if required to maintain a temperature between 15 and 25^{O}C.

Table 1. Products tested

MOPS code number	Biopesticide or conventional pesticide
Water control	-
Teppeki (flonicamid)	conventional
130	biopesticide
62	biopesticide
208	conventional
59	conventional
179	biopesticide
205	biopesticide

Plants were purchased as plugs and potted into Levington M2 Pot/Bedding Compost in 9cm diameter pots on 20th May. Twelve plants were arranged in four rows of three in each of 48 plots. Each plot was enclosed within a mesh cage (0.5m x 0.4m x 0.4m). Plants were watered from beneath using the capillary matting.

The population of whitefly used was established from a population of whitefly supplied by David Talbot (ADAS) from a commercial nursery. Each plot was infested with 50 adult whitefly on the 3rd July 2014 and then a further 30 adult whitefly introduced on the 17th July 2014.

An application rate for each plant protection product tested was agreed with the product manufacturers. All plant protection products were applied using an electric sprayer fitted with an HC/1.74/3 nozzle, in 600 litres of water per hectare using 3 bar pressure. A water control was applied using the same water volume and pressure. No adjuvants were used for any products tested. Each plant protection product and the water control was applied at weekly intervals for four weeks. The numbers of whitefly eggs, nymphs and adults on selected, marked leaves were recorded one day before the first spray application on the 1st August 2014 and then at three and six days after this application. Whitefly numbers were then recorded in exactly the same way six days after the second spray application (date of assessment = 8th August 2014), third (15th August 2014) and fourth (22nd August 2014) spray applications. A final assessment was made on the 19th September which was done by counting the numbers of adult whitefly caught on sticky traps placed in the cages. This was done 28 days after the final spray application. In addition, assessments of phytotoxicity were completed after each spray application.

Products 62 and 179 caused significant reductions in numbers of whitefly nymphs and products 62, 205 and 179 caused significant reductions in numbers of whitefly eggs, but this did not happen on every sampling occasion. All of the treatments reduced the numbers of whitefly adults caught on sticky traps 28 days after the final spray application, with the standard (Teppeki) and the products 208 and 59 reducing whitefly numbers close to zero in each plot (Figure 1).

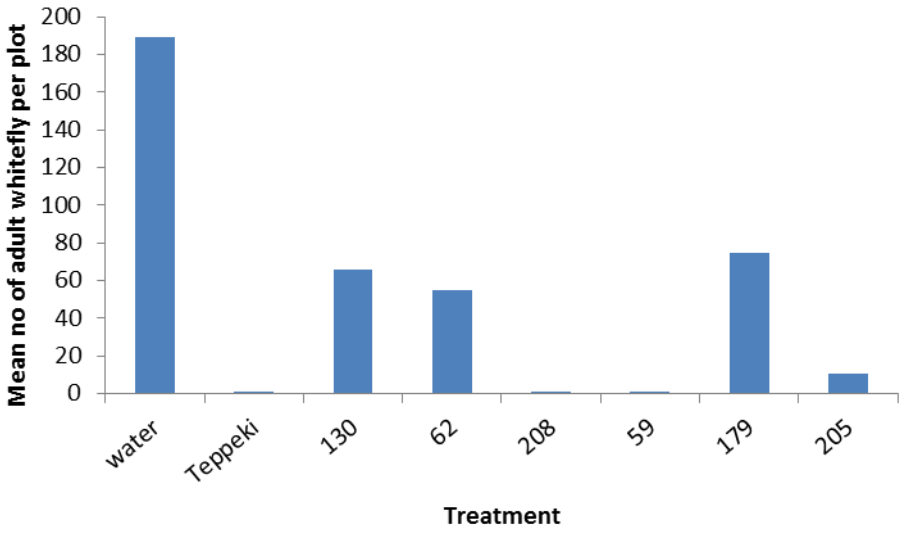


Figure 1. Mean numbers (backtransformed) of adult whitefly per plot collected on sticky traps 28 days after the final spray application.

There was no or limited phytotoxicity caused by any of the plant protection products tested. A very small number of leaves were observed with browning of the leaf edges and speckling of the flowers for some of the products tested.

Action Points

- A range of products have been identified which have potential as whitefly treatments. They all appear to have their main effect during the nymphal / pupal stages and preventing the emergence of adult whitefly from the pupae.
- Flonicamid (here applied as Teppeki, which is used for the control of aphids on wheat and potato) also effectively controlled glasshouse whitefly and therefore Mainman, an identical product which has an EAMU (0045 of 2013) for use on ornamentals, should also be effective.

Science Section

Introduction

Various whitefly species can infest ornamental plants but the most common in the UK is glasshouse whitefly, *Trialeurodes vaporariorum*. Common protected ornamental and HNS hosts include *Abelia*, *Abutilon*, *Ceanothus*, *Fuchsia*, *Pelargonium*, poinsettia, *Primula* and *Salvia spp.* Infested plants become contaminated with sticky honeydew excreted by whiteflies and this allows the growth of sooty moulds. In severe infestations, leaf yellowing and plant stunting occurs. The presence of whiteflies and damage symptoms can cause ornamental plants to be unmarketable.

Most UK glasshouse whitefly populations developed resistance to pyrethroid pesticides such as deltamethrin (e.g. Decis) by the mid 1980's (Wardlow, 1985).This pyrethroid and pyrethrum (e.g. Spruzit) resistance is still prevalent in current whitefly populations. Resistance later developed to the insect growth regulators buprofezin (Applaud) and teflubenzuron (Nemolt), (Gorman *et al*., 2002) but these pesticides are no longer available for whitefly control in the UK. Resistance to neonicotinoid insecticides has so far only been recorded in one glasshouse whitefly population in the UK, which was confirmed to be resistant to imidacloprid (Intercept 70 WG) on protected HNS (Gorman *et al*., 2007).

Due to these resistance problems, leading growers of protected ornamentals and HNS use biological control methods for glasshouse whitefly within IPM programmes. These mainly rely on the parasitoid *Encarsia formosa*, although there has also been some recent use of the predatory mite *Amblyseius swirskii* which feeds on both thrips larvae and whitefly eggs and young scales. However, IPM-compatible pesticides or biopesticides are often still needed within IPM for whitefly control, e.g. early season when temperatures are too low for optimal efficacy of biological control agents, or to supplement control during the summer when whitefly numbers can increase rapidly.

Pesticides commonly used by growers for whitefly control within IPM include pymetrozine (Chess WG), spiromesifen (Oberon) and flonicamid (Mainman). Other pesticides used include spirotetramat Movento) and the neonicotinoids acetamiprid (Gazelle SG) and thiacloprid (Calypso). However these products are less compatible with IPM and although these particular neonicotinoids are not included in the current restrictions on use of neonicotinoids, many retailers are asking growers not to use any neonicotinoids at all on their produce. This will further restrict the pesticide options for whitefly control. Biopesticides currently used for whitefly control include the plant extract product maltodextrin (Majestik) and the entomopathogenic fungus *Beauveria bassiana* (Naturalis-

L). However, control by Naturalis-L is not always fully effective in commercial glasshouses or tunnels, possibly due to humidity requirements following application.

Materials and methods

Site and crop details

Table 2. Test site and plot design information

Test location:	
County	Warwickshire
Postcode	CV35 9EF
Soil type/growing medium	Levington M2 pot/bedding compost
Nutrition	N/A
Crop	Verbena
Cultivar	Quartz
Glasshouse* or Field	Glasshouse
Date of planting/potting	Plug plants potted up on 20th May 2014
Pot size	9cm diameter pots
Number of plants per plot	12
Trial design (layout in Appendix C)	Two replicates (glasshouses) of an incomplete (4(3)*4)/2 Trojan square
Number of replicates	6 replicate plots per treatment
Plot size w (m), l (m), total area (m²)	50cm x 40cm x 40cm high ; 0.2m^2
Method of statistical analysis	Weekly counts were analysed using a multi-stratum ANOVA with plant position (bottom, middle, top) included as a sub-plot factor (mean values calculated across the five assessed plants to cope with missing leaves on some plants). The final sticky-trap count was analysed in a similar way, but without the sub-plot factor for position. Initial assessment of the residuals suggested that there was a variance-mean relationship for most of the variables to be analysed, so all variables were log-transformed prior to analysis. Back-

	transformed means were calculated for all tables of means, and individual t-tests calculated for comparisons of treatments with the untreated control.

*Temperature and relative humidity settings are given in Appendix B

Treatment details

Table 3. Detail of products tested

MOPS code number	Active ingredient(s)	Manufacturer	Batch number	% a.i	Formulation type
1. Water (-ve control)					
2. Teppeki (+ve control)	Flonicamid	Belchim Crop Protection	1612-05	500g/kg	WG
3. 130	Azadiractin A	Trifolio-M	140414A	1%	EC
4. 62	Terpenoid blend QRD 460	Bayer Crop Sciences	2014-004865	16.75%	OD
5. 208	Carboxylic acid and potassium salt	Alpha BioPesticides Ltd	ABP-617 (T134A)	-	-
6. 59	Sulfoxaflor	Dow Agrosciences	ENBK-143945-007A	120g/l	SC
7. 179	Orange oil	OroAgri	N/7579	60g/l	SL
8. 205	*Metarhizium anisopliae* var *anisolpliae* strain F52	Novozymes	1420 NFEE16	11%	EC

Table 4. Treatments

Product name or MOPS code number	Application timing	Product rate	Spray volume (L/ha)
1. Water (-ve control)	Weekly x 4	-	600
2. Teppeki (+ve control)	Weekly x 4	0.14kg/ha	600
3. 130	Weekly x 4	1.8l/ha	600
4. 62	Weekly x 4	3.9l/ha	600
5. 208	Weekly x 4	6l/ha	600
6. 59	Weekly x 4	0.4l/ha	600
7. 179	Weekly x 4	2.4l/ha	600
8. 205	Weekly x 4	1.25l/ha	600

Application timing	
A1	1st August 2014
A2	8th August 2014
A3	15th August 2014
A4	22nd August 2014

Table 5. Application details

Application No.	**A1**	**A2**	**A3**	**A4**
Application date	01/08/2014	08/08/2014	15/08/2014	22/08/2014
Time of day	12.30	12.00	11.30	11.30
Application method	An electric sprayer (CC25-N) fitted with a HC/1.74/3 nozzle, in 600 litres of water per ha using 3 bar pressure	An electric sprayer (CC25-N) fitted with a HC/1.74/3 nozzle, in 600 litres of water per ha using 3 bar pressure	An electric sprayer (CC25-N) fitted with a HC/1.74/3 nozzle, in 600 litres of water per ha using 3 bar pressure	An electric sprayer (CC25-N) fitted with a HC/1.74/3 nozzle, in 600 litres of water per ha using 3 bar pressure
Temperature of air – max/min (°C)	24.5	29.0	23.8	20.6
Relative humidity	54	55	53	50

(%)				
Cloud cover (%)	N/A	N/A	N/A	N/A
Crop growth stage	Flowering	Flowering	Flowering	Flowering
Crop comments	-	-	-	-
Other*:	-	-	-	-

*Includes soil temperature and moisture details where relevant

The application method was agreed following consultation with industry representatives, a spray application expert (David Talbot, ADAS) and product manufacturers. Water-sensitive paper was used to provide an assessment of the spray coverage on the plants. Papers were attached to a group of Verbena plants that were set out on the glasshouse bench in the same arrangement used in the experimental plots. The plants were then sprayed with water, and the spray coverage on the upper and lower surfaces of the papers were assessed, mimicking spray application to upper and lower leaf surfaces. Assessments were done by noting presence / absence of water marks on the paper. An additional test was done to confirm the flow of product 205 with the spray apparatus. Samples were taken from the tank before spraying and spray samples collected both with the filter in the nozzle and with the filter removed. Spores in the tank and spray samples were enumerated using an improved Neubauer haemacytometer. There was no statistical difference between the number of spores sprayed in the presence or absence of the nozzle filter and the tank (data not shown).

Target pest

Table 6. Target pest

Common name	Scientific Name	Infection level pre-application
Glasshouse whitefly	*Trialeurodes vaporariorum*	ca. 20 nymphs per marked leaf

Each plot was infested with 50 adult whitefly on the 3rd July 2014 and then a further 30 adult whitefly introduced on the 17th July 2014. The whitefly used to infest the trial were collected from a stock culture at Warwick Crop Centre which were maintained on Verbena plants within a controlled environment room and which originated from an infested commercial nursery, courtesy of David Talbot (ADAS).

Assessments

A pre-assessment of the plant identified three heights (top, middle and bottom) on five plants within each plot with similar whitefly numbers. These leaves were marked. For each assessment, the marked leaves were assessed, using a hand lens, for eggs, scales and adults. Sticky traps were placed in the cages after the final assessment and counted three weeks later.

Table 7. Assessments

Assessment No.	Date	Growth stage	Time of assessment relative to last application	Time of assessment relative to the first application	Assessment type(s) (e.g. no./% LAI/crop safety)
1	29-30/07/2014	Flowering	1-2 days before first application	-	Whitefly count
2	04/08/2014	Flowering	3 days post application A1	3 d	Whitefly count & crop safety
3	07/08/2014	Flowering	6 days post application A1	6d	Whitefly count & crop safety
4	14/08/2014	Flowering	6 days post application A2	13d	Whitefly count & crop safety
5	21/08/2014	Flowering	6 days post application A3	20d	Whitefly count & crop safety
6	28/08/2014	Flowering	6 days post application A4	27d	Whitefly count & crop safety
7	19/09/2014	Senescing	28 days post application A4	48d	Whitefly count

Statistical analysis

Weekly counts were analysed using a multi-stratum ANOVA with plant position (bottom, middle, top) included as a sub-plot factor (mean values calculated across the five assessed plants to cope with missing leaves on some plants). This analysis also considered any interaction between treatment and position. Total counts of all life stages, and combined counts of adults and nymphs were also analysed. The final sticky-trap count was analysed in a similar way, but without the sub-

plot factor for position. Initial assessment of the residuals suggested that there was a variance-mean relationship for most of the variables to be analysed, so all variables were log-transformed (log_e) prior to analysis. Back-transformed means were calculated for all tables of means, and individual t-tests calculated for comparisons of treatments with the untreated control.

Results

Spray coverage

The application method used achieved good spray coverage on the upper leaf surfaces in the upper and middle crop canopies. However, spray coverage on the lower leaf surface was poor at all positions within the crop canopy in particular in the middle and lower canopies (Figure 2).

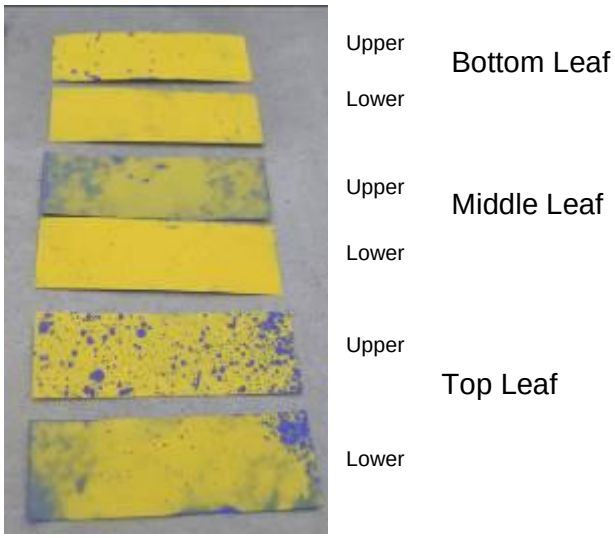


Figure 2. Spray coverage on water sensitive paper positioned on the upper and lower leaf surfaces in the upper, middle and lower crop canopy.

Control of Whitefly

Results are summarised in Tables 8, 9, 10 and 11 (with ANOVA statistics) and Figure 3, 4, 5 and 6 below.

Control of adult whitefly caught on sticky traps at the end of the experiment.
All of the treatments significantly ($p<0.001$) reduced the numbers of adult whitefly caught on sticky traps compared to the untreated water control. In particular, the products Teppeki, 208, 59 and

205 reduced whitefly numbers close to zero in each plot by the end of the experimental period (Table 8, Figure 3).

Table 8. Effect of treatments on numbers of adult whitefly caught on sticky traps at the end of the experiment. Data presented as mean number of whitefly per plot (log_e transformed). Numbers with an * are significantly different at p<0.001 from the untreated water control based on individual contrasts (sed).

Product name or MOPS code	Mean no. whitefly per plot (log_e transformed) assessed on 19/09/2014
1. Water (-ve control)	5.242
2. Teppeki (+ve control)	0.300*
3. 130	4.192*
4. 62	4.008*
5. 208	-1.407*
6. 59	-0.612*
7. 179	4.313*
8. 205	2.348*
F value (7 df)	0.359
sed	0.7673

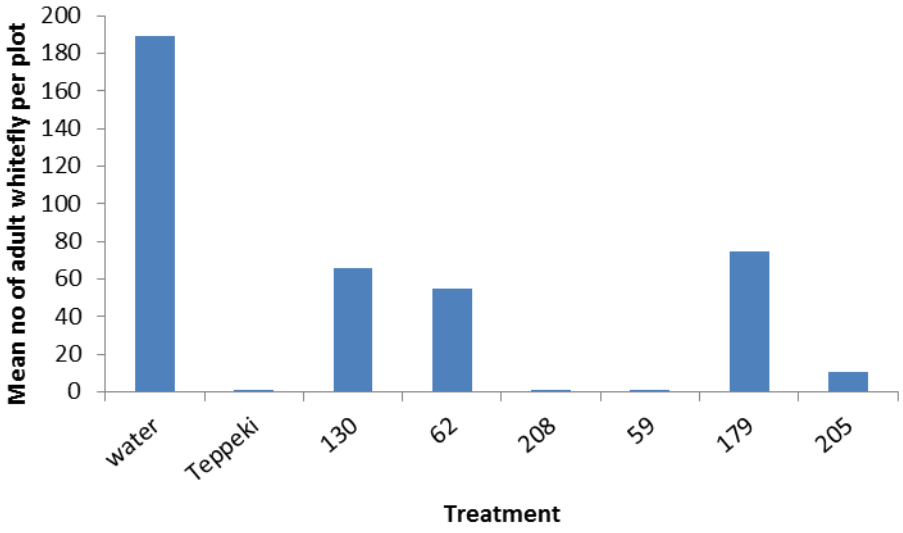


Figure 3. Mean numbers (backtransformed) of adult whitefly per plot collected on sticky traps 21 days after the final spray application.

Effect of treatments on numbers of adult whitefly counted on marked leaves during the experiment. Overall there was little effect of treatment on numbers of whitefly adults counted on marked leaves during the experiment (Table 9, Figure 4). Statistical analysis showed that there was no statistically significant difference in whitefly adult numbers before or after the first spray. The second spray of product 179 and the third spray of product 130 significantly ($p<0.001$) reduced the number of adults compared to the water control. Six days after the final spray there was a significant reduction ($p<0.001$) in whitefly adults observed in plots treated with products 130, 179 and 205. These results should be interpreted with caution because the counts of the adult whitefly on marked leaves are not thought to be reliable. The adults would often fly off the marked leaves when examined, probably as a result of disturbance. Moreover, because the adult whitefly are very mobile, there is no guarantee that any adult whitefly that were counted on a marked leaf on any particular assessment day originated from an egg laid on that leaf. Hence, counting adult whitefly on marked leaves is unlikely to be an accurate estimate of the adult whitefly population in the whole plot.

Table 9. Effect of treatments on whitefly adults. Data presented as mean number of whitefly per leaf (log_e transformed). Numbers with an * are significantly different at $p<0.001$ from the untreated water control based on individual contrasts (sed).

Product name or MOPS code	30/07/2014	04/08/2014	07/08/2014	14/08/2014	21/08/2014	28/08/2014
1. Water (-ve control)	-1.2476	-0.8139	-0.974	-1.068	-1.136	-1.483
2. Teppeki (+ve control)	-0.5080	-0.5189	-1.098	-1.153	-1.655	-1.931
3. 130	-1.0364	-0.8564	-1.112	-1.485	-1.802*	-2.017*
4. 62	-1.0415	-0.8680	-1.287	-0.902	-1.059	-1.590
5. 208	-1.1376	-0.7996	-1.159	-1.272	-1.454	-1.698
6. 59	-0.6637	-1.1471	-0.932	-1.451	-1.236	-1.263
7. 179	-0.9679	-1.1120	-0.985	-1.761*	-1.660	-1.998*

8. 205	-1.1540	-0.9465	-1.194	-1.291	-0.979	-1.976*
F value (7 df)	0.782	0.865	0.996	0.629	0.544	0.332
sed	0.4703	0.4268	0.4934	0.4338	0.4668	0.3647

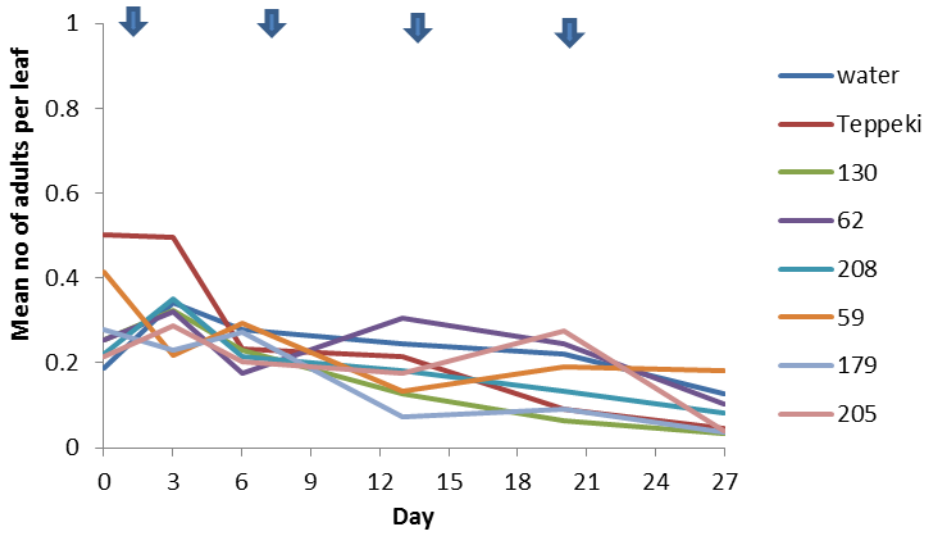


Figure 4. Mean number (backtransformed) of whitefly adults per leaf on each assessment date (5 plants at 3 canopy heights) sampled in each plot. The arrows represent the dates of treatment application.

Effect of treatments on numbers of whitefly nymphs counted on marked leaves.

After the first spray application only products 62 and 179 showed a significant ($p < 0.001$) reduction in numbers of whitefly nymphs counted on marked leaves compared to the untreated water control (Table 10, Figure 5). In addition, product 62 showed a significant reduction three days after the first spray application and also six days after the second application. Six days after the final spray application only product 179 had a significant reduction in whitefly nymphs counted compared to the untreated water control.

Table 10. Effect of treatments on whitefly nymphs. Data presented as mean number of whitefly per leaf (log_e transformed). Numbers with an * are significantly different at $p<0.001$ from the untreated water control based on individual contrasts (sed).

Product name or MOPS code	30/07/2014	04/08/2014	07/08/2014	14/08/2014	21/08/2014	28/08/2014
1. Water (-ve control)	1.804	2.225	2.586	2.311	0.888	1.802

2. Teppeki (+ve control)	2.212	2.515	2.370	2.290	1.688	1.706
3. 130	2.257	2.124	2.109	1.947	1.189	1.685
4. 62	1.774	1.156*	1.861*	1.726*	1.623	1.878
5. 208	1.601	2.051	2.437	2.338	1.219	1.676
6. 59	1.974	2.211	2.058	2.156	1.336	2.165
7. 179	1.966	2.192	1.900*	1.852	0.828	1.237*
8. 205	1.799	2.020	2.092	1.932	1.535	1.650
F value (7 df)	0.940	0.064	0.745	0.636	0.689	0.494
sed	0.5570	0.3757	0.4657	0.3761	0.5446	0.3800

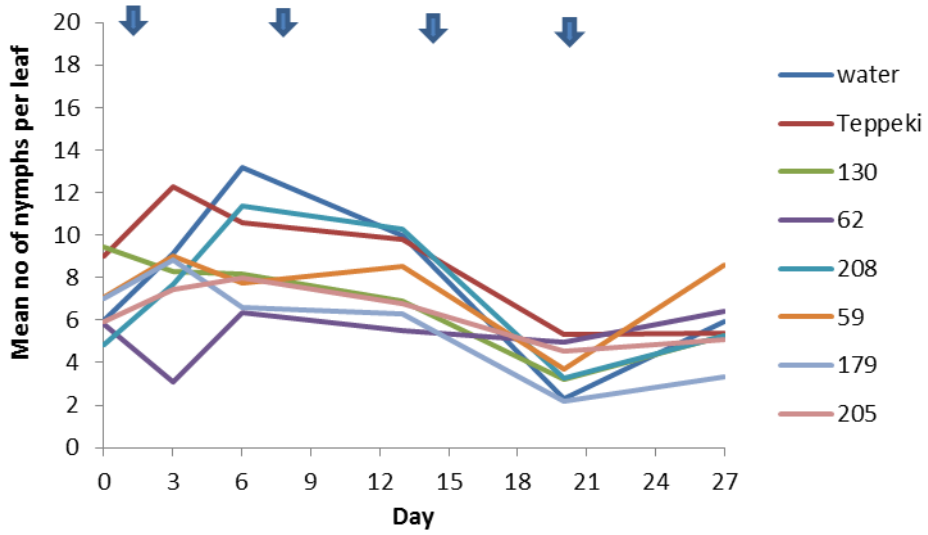


Figure 5. Mean number (backtransformed) of whitefly nymphs per leaf on each assessment date (5 plants at 3 canopy heights) sampled in each plot. The arrows represent the dates of treatment application.

Effect of treatments on numbers of whitefly eggs counted on marked leaves.

Three of the treatments showed a significant ($p<0.001$) reduction in whitefly eggs compared to the untreated water control during the experiment (Table 11, Figure 6). Three days after the first spray application products 62 and 205 showed a significant reduction in whitefly eggs compared to the untreated water control. There was no significant difference in egg number observed with any of the treatments observed six days after the first application. In addition, product 62 showed a

significant reduction in whitefly eggs compared to the untreated water control six days after the third spray application. Numbers of whitefly eggs were also significantly reduced six days after the second spray with product 179 compared to the untreated water control. It is not known whether reductions in numbers of eggs was caused by egg mortality, a decrease in the rate of oviposition by adult females, or a combination of both factors.

Table 11. Effect of treatments on whitefly eggs. Data presented as mean number of whitefly per leaf (log_e transformed). Numbers with an * are significantly different at p<0.001 from the untreated water control based on individual contrasts (sed).

Product name or MOPS code	30/07/2014	04/08/2014	07/08/2014	14/08/2014	21/08/2014	28/08/2014
1. Water (-ve control)	-0.0595	-0.0967	0.1975	-0.9723	-0.826	-0.7964
2. Teppeki (+ve control)	0.5274	0.1071	-0.6258	-0.8963	-0.994	-1.3159
3. 130	1.1180	-0.1345	-0.4908	-0.5490	-1.128	-1.5229
4. 62	0.0021	-0.7921*	-0.1883	-1.2731	-2.102*	-0.7209
5. 208	0.0707	-0.0550	-0.5529	0.1466	-1.400	-0.5484
6. 59	1.0425	0.3843	-0.4806	0.0898	-1.441	-0.5337
7. 179	0.5119	0.4957	-0.4204	-2.2135*	-1.626	-1.1021
8. 205	-0.1856	-1.0994*	0.2947	-0.6029	-0.280	-0.8106
F value (7 df)	0.452	0.034	0.982	0.271	0.447	0.861
sed	0.701	0.472	1.1079	0.943	0.798	0.757

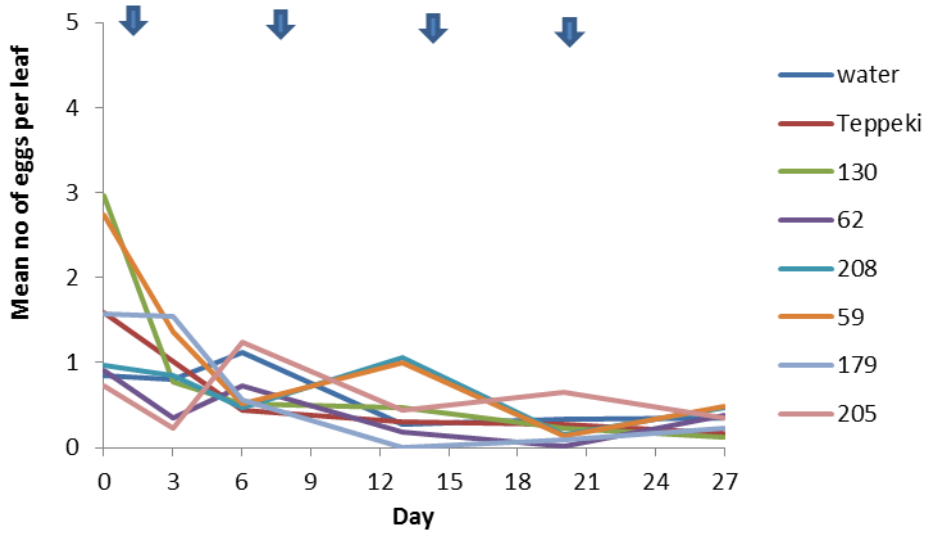


Figure 6. Mean number (backtransformed) of whitefly eggs per leaf on each assessment date (5 plants at 3 canopy heights) sampled in each plot. The arrows represent the dates of treatment application.

Crop damage

Crop damage (associated with the application of treatments, i.e. phytotoxicity) was recorded as the number of damaged leaves and flowers in all plots and severity of damage (none, slight, moderate and high). All phytotoxic damage observed consisted of changes in leaf or flower colour, specifically there was slight browning of the leaves and pale spots on the flowers for products 130, 208, 59 and 179 on some occasions. The numbers of leaves affected in this way were very small and were not considered to be high enough to warrant statistical analysis. Each plot had about 120 leaves. The highest damage was recorded for products 208 and 59 where 2 leaves per plot showed slight damage.

Table 12. Effect of treatments – crop damage. Data presented as the number of leaves and flowers damaged per plot.

Product name or MOPS code	04/08/2014	07/08/2014	14/08/2014	21/08/2014	28/08/2014
1. Water (-ve control)	0 leaves 0 flowers	0 leaves 0 flowers	0 leaves 0 flowers	0 leaves 0 flowers	0 leaves 0 flowers
2. Teppeki (+ve control)	0 leaves 0 flowers	0 leaves 0 flowers	0 leaves 0 flowers	0 leaves 0 flowers	0 leaves 0 flowers

3. 130	0 leaves 0 flowers	0 leaves 0 flowers	0 leaves 0 flowers	0 leaves 1 flower (slight)	0 leaves 0 flowers
4. 62	0 leaves 0 flowers	0 leaves 0 flowers	0 leaves 0 flowers	0 leaves 0 flowers	0 leaves 0 flowers
5. 208	0 leaves 0 flowers	0 leaves 0 flowers	1 leaf (slight) 0 flowers	1 leaf (slight) 1 flower (slight)	2 leaves (slight) 1 flower (slight)
6. 59	0 leaves 0 flowers	0 leaves 0 flowers	0 leaves 0 flowers	1 leaf (slight) 0 flowers	2 leaves (slight) 0 flowers
7. 179	0 leaves 0 flowers	0 leaves 0 flowers	0 leaves 1 flower (slight)	0 leaves 3 flowers (slight)	0 leaves 3 flowers (slight)
8. 205	0 leaves 0 flowers	0 leaves 0 flowers	0 leaves 0 flowers	0 leaves 0 flowers	0 leaves 0 flowers

Formulations

No problems were encountered during mixing or application of any of the product formulations under test.

Effect on non-target

No effect on other pests was noted during the completion of this trial. An infestation of western flower thrips was recorded and the treatments appeared to have little effect in reducing their numbers.

Discussion

All the products evaluated in this study caused significant reductions in the numbers of adult whitefly counted on sticky traps at the end of the experiment, as compared to the control. The sticky trap counts provide an assessment of the efficacy of a treatment against the whitefly population as a whole within a cage. We believe that this is the most reliable estimate of product efficacy for the whole experiment. Biopesticide products 130, 179 and 205 caused significant reductions in numbers of whitefly adults on marked leaves on some occasions during the experiment, but the other products had no effect on numbers of adults. Biopesticide products 62 and 179 caused significant reductions in numbers of whitefly nymphs on marked leaves on some occasions during the experiment, but the other products had no effect on numbers of nymphs. Biopesticide products 62, 205 and 179 caused significant reductions in numbers of eggs on some assessment dates. As not all products affected eggs or nymphs, this suggests that the products

caused the whitefly to die mainly in the pupal stage, either by killing them outright or by making them too weak to emerge from the puparium.

The slow speed of kill is reflected in what is known about the mode of action of the different products. Treatment 2 (Teppeki, the positive control) and product 130 are feeding inhibitors and so might be expected to take some days to kill whitefly nymphs. Product 130 is also reported to coat the outside of insects and prevent them from acquiring oxygen, which again suggest that death is unlikely to be instantaneous. Product 62 has contact action and is reported to have activity against eggs, nymphs and adults, and work by acting as a feeding inhibitor, degrading cuticle, and destroying the lining of the trachea. Again this suggests that death is unlikely to be immediate. Conventional product 59 is reported to be an insect neurotoxin with contact and stomach action, and to be systemic. These attributes might be expected to give it a higher efficacy than some other products. Product 205 is a microorganism that infects insects by growing on the cuticle, and the time taken to achieve this means that death is not instantaneous.

The products tested here have potential for whitefly control, but we believe they are unlikely to be used as stand-alone “magic bullet” treatments, but rather they will have more value as supplementary treatments to back up biocontrol with parasitoids and predators in an IPM programme. If most of the products act on the pupal stage, then they would complement biocontrols that act on other life stages, for example parasitoids such as *Encarsia formosa* that parasitize larval stages and predatory mites such as *Amblyseius swirskii* that feed on eggs and very young larval stages.

For the experiment, the whitefly population in each plot was established over the course of a month prior to the first spray application. Glasshouse whitefly has a development time, from egg to adult, of about 25 days under typical glasshouse conditions (the exact time to development is dependent on the temperature and ranges from about 18 – 31 days). Therefore, the whitefly population would have consisted of a mixture of different development stages (eggs, nymphs, pupae and adults) within each plot during the experiment. Adult whiteflies tend to lay their eggs in the upper part of the canopy, and move upwards to oviposit as the plant grows. Oviposition occurs over a finite period, meaning that the population of whitefly on the same leaf tends to consist of similar developmental stages. The time over which egg laying occurs on a particular leaf is likely to vary with factors such as plant species, leaf size etc. However – with the exception of the motile adult stages - it is likely that the population of whitefly on the marked leaves evaluated in this experiment were at similar development stages in different plots. This would explain why the numbers of nymphs counted on the marked leaves declined in a fairly synchronized way over time for all treatments, with a more sudden drop in numbers at day 20; this pattern would be consistent with the metamorphosis of nymphs into adults which would then disperse into the plot. The adult stages are highly motile, they move up the plant as it grows in order to lay eggs, and we noticed that they flew off leaves as soon

as the cages were removed from the plots in order to assess the plants. Therefore, the counts of numbers of adult whitefly on the marked leaves are unlikely to be related to the numbers of eggs and nymphs of the same cohort on marked leaves. Combined with the fact that all of the treatments tested were relatively slow acting, this would explain why overall there was little effect of any treatment on numbers of whitefly adults counted on marked leaves during the experiment.

There was generally poor spray coverage on the underside of leaves, particularly for the mid part of the plant where the whitefly populations were largest. This is likely to have affected the efficacy of all treatments. Our spray applications were designed to mimic those done by commercial growers, but given the small plot sizes it is likely that we achieved better spray coverage than would be likely in commercial nurseries. This indicates the importance of developing new, more effective methods of application for these new types of product, and in particular to get good coverage onto the underside of leaves, for example by using electrostatically charged spray droplets or by using mists.

On ornamentals, growers often want a rapid knock down of adults, for example just before sale. There was no evidence of a direct effect of any treatment on adults in this study; however – as described above – the design of the experiment was not conducive to accurately measuring direct effects on adults. The best way to do this would probably be as a laboratory based bioassay in which a fixed age population of adult whitefly is given a spray treatment and then its survival monitored over time. However this would still leave an issue of the importance of obtaining good spray coverage on the underside of leaves for those products with contact action.

Treatments were applied four times at weekly intervals, but for some products this is a higher frequency of applications than that on the product label. For example, treatment 2 (Teppeki), has a maximum of three applications per year, with a maximum of two in succession in order to reduce the risk of resistance developing. Additionally, product 59 was applied at a lower than recommended dose, as advised by the manufacturer, to accommodate this. The programme of repeated sprays used here was done in order to be able to identify effective treatments and rank them in order of efficacy, but for some products using a series of weekly sprays may not be practical or desirable.

Conclusions

- All of the treatments that were tested caused significant reductions in the number of adult whitefly counted on sticky traps at the end of the experiment.
- Products 62 and 179 caused significant reductions in numbers of whitefly nymphs on marked leaves on some occasions during the experiment.

- Products 62, 205 and 179 caused significant reductions in numbers of whitefly eggs on marked leaves on some occasions during the experiment.
- Some phytotoxicity symptoms, seen as browning along leaf margins and speckling on flowers were recorded but this was considered ‘slight’ in terms of severity and affected relatively few leaves and flowers.

References

Gorman, K., Devine, G., Bennison, J., Coussons, P., Punchard, N. & Denholm, I. (2007). Report of resistance to the neonicotinoid insecticide imidacloprid in *Trialeurodes vaporariorum* (Hemiptera: Aleyrodidae). *Pest Management Science* **63**, 555-558.

Gorman, K., Hewitt, F., Devine, G. & Denholm, I. (2002). New developments in insecticide resistance in the glasshouse whitefly (*Trialeurodes vaporariorum*) and the two-spotted spider mite (*Tetranychus urticae*) in the UK. *Pest Management Science* **58**, 123-130.

Wardlow, L.R. (1985). Pyrethroid resistance in glasshouse whitefly (*Trialeurodes vaporariorum* (Westwood). *Med. Fac. Landbouw, Rijksuniv, Gent* **50**, 164-165.

Appendix A – Study conduct

Warwick Crop Centre are officially recognised by United Kingdom Chemical Regulations Directorate as competent to carry out efficacy testing in the categories of agriculture, horticulture, stored crops, biologicals & semiochemicals. National regulatory guidelines were followed for the study.

GLP compliance will not be claimed in respect of this study.

Relevant EPPO/CEB guideline(s)		**Variation from EPPO**
PP 1/152(3)	Design and analysis of efficacy evaluation trials	
PP 1/135(3)	Phytotoxicity assessment	
PP 1/181(3)	Conduct and reporting of efficacy evaluation trials including GEP	
PP 1/36(3)	Efficacy evaluation of insecticides: *Whiteflies (Trialeurodes vaporariorum, Bemisia tabaci)* on protected crops	Verbena is not listed as a test crop. Separate glasshouse compartments were not used for different treatments. Six replicates of each treatment.

There were no significant deviations from the EPPO and national guidelines.

Appendix B – Meteorological data

Location of the weather station	52 12 18^{O} N; 1 36 00^{O}W
Distance to the trial site	350m
Origin of the weather data	Warwick Crop Centre met station

Long-term averages from *location*			
Month/period	**Min temp (°C)**	**Max temp (°C)**	**Rainfall (mm)**
July 1959 -2013	11.54	22.46	2.05
August 1959-2013	11.93	22.11	1.79
September 1959-2013	8.99	18.61	1.55

Average conditions during the trial (datalogger within glasshouse compartment)

Month/period	**Av temp (°C)**	**Min temp (°C)**	**Max temp (°C)**	**Av RH (%)***	**Rainfall (mm)**
July 2013	22.43	13.8	35.3	55	-
August 2013	20.44	14.0	37.0	62	-
September 2013	20.44	10.4	31.9	58	-

***protected crops only**

Weather at treatment application: (datalogger within glasshouse compartment)

Month/period	**Min temp (°C)**	**Max temp (°C)**	**Rainfall (mm)**
01/08/2014	17.6	30.4	-
08/08/2014	17.6	35.1	-
15/08/2014	16.2	28.1	-
22/08/2014	17.4	28.1	-

Appendix C – Agronomic details

Growing system

Crop	Cultivar	Planting/sowing date	Row width (m) or pot spacing
Verbena	Quartz	Plug plants potted up on 20th May 2014	Pots arranged in 4 rows of three – spacing between pots 2.5cm

Other pesticides - active ingredient(s) / fertiliser(s) applied to the trial area

Date	Product	Rate	Unit

Details of irrigation regime (pot-grown crops)

Type of irrigation system employed (e.g. overhead sprinkler, hand watering, drip, ebb and flow, capillary sandbed or capillary matting)
Drip irrigation onto capillary matting

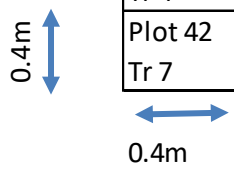
Appendix D – Trial layout

Glasshouse 1

Plot 1 Tr 1	Plot 3 Tr 6	Plot 5 Tr 8	Plot 7 Tr 5
Plot 2 Tr 7	Plot 4 Tr 4	Plot 6 Tr 2	Plot 8 Tr 3
Plot 9 Tr 3	Plot 11 Tr 5	Plot 13 Tr 7	Plot 15 Tr 6
Plot 10 Tr 8	Plot 12 Tr 2	Plot 14 Tr 4	Plot 16 Tr 1
Plot 17 Tr 5	Plot 19 Tr 1	Plot 21 Tr 3	Plot 23 Tr 2
Plot 18 Tr 4	Plot 20 Tr 8	Plot 22 Tr 6	Plot 24 Tr 7

Glasshouse 2

Plot 25 Tr 3	Plot 27 Tr 4	Plot 29 Tr 1	Plot 31 Tr 2
Plot 26 Tr 6	Plot 28 Tr 5	Plot 30 Tr 8	Plot 32 Tr 7
Plot 33 Tr 8	Plot 35 Tr 1	Plot 37 Tr 6	Plot 39 Tr 5
Plot 34 Tr 2	Plot 36 Tr 7	Plot 38 Tr 4	Plot 40 Tr 3
Plot 41 Tr 4	Plot 43 Tr 8	Plot 45 Tr 5	Plot 47 Tr 1
Plot 42 Tr 7	Plot 44 Tr 3	Plot 46 Tr 2	Plot 48 Tr 6



Appendix E – Copy of the Certificate of Official Recognition of Efficacy Testing Facility or Organisation

Certificate of

Official Recognition of Efficacy Testing Facilities or Organisations in the United Kingdom

This certifies that

Warwick Crop Centre, School of Life Sciences

complies with the minimum standards laid down in Regulation (EC) 1107/2009 for efficacy testing.

The above Facility/Organisation has been officially recognised as being competent to carry out efficacy trials/tests in the United Kingdom in the following categories:

Agriculture/Horticulture

Date of issue: 14 May 2012
Effective date: 20 March 2012
Expiry date: 19 March 2017

Signature *Authorised signatory*

Certification Number
ORETO 299

HSE
Chemicals Regulation Directorate

Department of Agriculture and Rural Development

Appendix F – Photographs

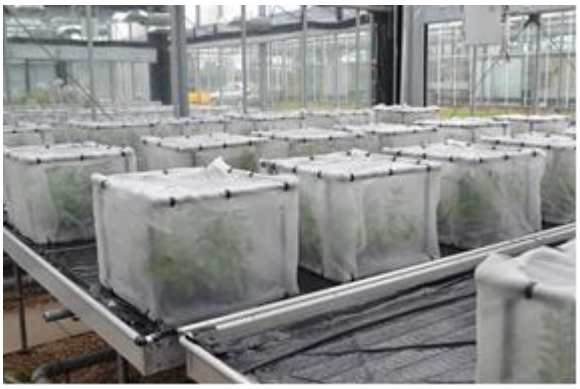


Figure 7. Arrangement of glasshouse compartment

Figure 8. Arrangement of 12 verbena plants potted into 9 cm pots within a plot

Figure 9. Phytotoxicity damage to a flower treated with product 179

Figure 10. Phytotoxicity damage to a flower treated with product 208

