

# SCEPTREPLUS

## Final Trial Report

<b>Trial code:</b>	SP 56
<b>Title:</b>	Aphid control on containerised hardy nursery stock
<b>Crop</b>	Containerised hardy nursery stock ( <i>Hebe</i> ), trial data also applicable to other containerised protected or outdoor ornamentals.
<b>Target</b>	<i>Aphis gossypii</i>
<b>Lead researcher:</b>	Jude Bennison
<b>Organisation:</b>	ADAS
<b>Period:</b>	March to November 2020
<b>Report date:</b>	30 November 2020
<b>Report author:</b>	Elysia Bartel and Jude Bennison
<b>ORETO Number: (certificate should be attached)</b>	409

I the undersigned, hereby declare that the work was performed according to the procedures herein described and that this report is an accurate and faithful record of the results obtained

30/11/20

Date



Authors signature

# Trial Summary

## Introduction

The melon and cotton aphid (*Aphis gossypii*) is one of the most serious pest species in hardy nursery stock (HNS) due to its wide host plant range and resistance to several pesticide groups. Many growers use Integrated Pest Management programmes including aphid parasitoids and predators to provide aphid control. However, plant protection products can be necessary to control fast growing populations and to clean up pests on produce prior to dispatch. This experiment tested novel products for efficacy against *A. gossypii* in order to identify potential products suitable for inclusion in IPM programmes. Additional information was gained on efficacy against a natural infestation of peach-potato aphid, *Myzus persicae* (another pest of HNS with resistance to several actives).

## Methods

*Hebe* (cv. Purple Pixie) plants were grown in aphid-proof 'BugDorms' in a polytunnel at ADAS Boxworth from September to October 2020. Plants were infested with *Aphis gossypii* nine days before the first spray application. There were seven replicate cages per treatment and seven treatments applied (untreated, industry standard – Mainman, four biopesticides and one conventional insecticide). All treatments were applied using an Oxford precision sprayer in 600 L / ha water. Treatments were applied at rates and timings recommended by manufacturers, which varied from twice at 7-day intervals to three times at 5-day intervals. Assessment of numbers of *A. gossypii* and numbers of *Myzus persicae* (due to natural infestation) were made two days before the first application and three, six, 13 and 20 days after the first application.

## Results

Back-transformed mean numbers of <i>Aphis gossypii</i> per plant					
Date	09.09.20 (Day-2)	14.09.20 (Day 3)	17.09.20 (Day 6)	24.09.20 (Day 13)	01.10.20 (Day 20)
Treatment					
Untreated	13.89	10.46	10.44	12.08	11.32
Mainman	13.76	9.33	7.66	8.37	10.32
AHDB9968	13.83	8.86	3.42*	2.43*	3.29*
AHDB9921	14.00	11.19	12.24	5.27	2.88*
AHDB9920	13.86	8.25	7.36	8.67	9.43
AHDB9919	13.49	10.09	7.63	10.48	10.85
AHDB9918	13.93	10.25	10.55	14.49	14.46
	Not significantly different from untreated control (P>0.05)				
	*Significantly different from untreated control (P<0.05)				

## Conclusions

- Mainman, used as a single application as the industry standard positive control did not give significant reductions in either *A. gossypii* or *M. persicae* compared with untreated controls on any assessment date.
- The bioprotectant AHDB9968, applied three times at 5-day intervals was effective against *A. gossypii* and *M. persicae* from six days after the first application. The conventional insecticide AHDB9921, applied twice at 7-day intervals was effective against *A. gossypii* after 20 days and against *M. persicae* after 13 days from the first application. The bioprotectants AHDB9920 and AHDB9919, applied three times at 5-day intervals were ineffective against *A. gossypii* but were effective against *M. persicae* on one assessment date, 13 days after the first application.
- The bioprotectant AHDB9918, applied three times at 5-day intervals was ineffective against *A. gossypii* and *M. persicae*.
- No treatments caused phytotoxic effects on the tested plant species, *Hebe* var. Purple Pixie

**Take home message:**

If approved for use in ornamental plant production, one bioprotectant could be very useful for aphid control in an IPM programme. One conventional insecticide could also be useful for slower acting control of *A. gossypii* and *M. persicae* and two other bioprotectants could be useful against *M. persicae*.

## Objectives

1. To evaluate the effectiveness of new conventional insecticides and biopesticides against *Aphis gossypii* on protected hardy nursery stock as measured by numbers of live aphids per plant.
2. To monitor the treated plants (*Hebe* cv. Purple Pixie) for phytotoxicity.

## Trial conduct

UK regulatory guidelines were followed but EPPO guidelines took precedence. The following EPPO guidelines were followed:

Relevant EPPO guideline(s)		Variation from EPPO
PP1/023(2)	Aphids on ornamental plants	Species selected is <i>Hebe</i> . The trial will only be carried out in one growing season Plot size is < 2m <sup>2</sup> . Number of pots per plot is < 10.
PP1/135(4)	Phytotoxicity assessment	None
PP1/152(4)	Guideline on design and analysis of efficacy evaluation trials	None
PP1/225 (2)	Minimum effective dose	None
PP1/181 (4)	Conduct and reporting of efficacy evaluation trials including good experimental practice	None
PP 1/214(3)	Principles of acceptable efficacy	None
PP 1/224(2)	Principles of efficacy evaluation for minor uses	None

## Test site

Item	Details
Location address	Poytunnel 6, ADAS Boxworth, Battlegate Road, Boxworth, Cambridgeshire, CB23 4NN
Crop	<i>Hebe</i>
Cultivar	Purple Pixie
Soil or substrate type	Levington Container Nursery Stock general compost with Osmocote Exact standard 12 – 14m at 3kg / m <sup>3</sup> .
Agronomic practice	See Appendix
Prior history of site	Polytunnels used for horticultural and agricultural research

## Trial design

Item	Details
Trial design:	Randomised block
Number of replicates:	7
Row spacing:	9cm pots in two rows of 3 at 0.1 m spacing
Plot size: (w x l)	0.48 m x 0.48 m thrips-proof BugDorm cage
Plot size: (m <sup>2</sup> )	0.23 m <sup>2</sup>
Number of plants per plot:	6
Leaf Wall Area calculations	N/A

## Treatment details

AHDB Code	Active substance	Product name/ manufacturer s code	Formulation batch number	Content of active substance in product	Formulation type	Adjuvant
Untreated						
Mainman	Flonicamid	Mainman	9625-01	500 g / kg	WG	None
AHDB9968	N/D	N/D	N/D	N/D	N/D	None
AHDB9921	N/D	N/D	N/D	N/D	N/D	None
AHDB9920	N/D	N/D	N/D	N/D	N/D	None
AHDB9919	N/D	N/D	N/D	N/D	N/D	None
AHDB9918	N/D	N/D	N/D	N/D	N/D	None

Rainwater was used to apply AHDB9920 and AHDB9918.

## Application schedule

Treatment number	Treatment: product name or AHDB code	Rate of active substance (ml or g a.s./ha)	Rate of product (l or kg/ha)	Application code
1	Untreated			
2	Mainman	500 g / kg	0.14 kg / ha (EAMU 2013/0045)	A
3	AHDB9968	3-4%	3 L / ha	A, B, D
4	AHDB9921	N/D	0.25 L / ha	A, C
5	AHDB9920	51%	12 L / ha	A, B, D
6	AHDB9919	476 g / L	12 L / ha	A, B, D
7	AHDB9918	<50%	7 L / ha	A, B, D

## Application details

	Application A	Application B	Application C	Application D
Application date	11/09/20	16/09/20	18/09/20	21/09/20
Time of day	09:45	9:05	11:40	10:00
Crop growth stage (Max, min average BBCH)	(flower senescence, inflorescence emerging, flowering)	(flower senescence, inflorescence emerging, flowering)	(flower senescence, inflorescence emerging, flowering)	(flower senescence, inflorescence emerging, flowering)
Crop height (cm)	15	15	15	15
Crop coverage (%)	N/A	N/A	N/A	N/A
Application Method	Spray	Spray	Spray	Spray
Application Placement	Foliar	Foliar	Foliar	Foliar
Application equipment	Oxford Precision Sprayer (knapsack)	Oxford Precision Sprayer (knapsack)	Oxford Precision Sprayer (knapsack)	Oxford Precision Sprayer (knapsack)
Nozzle pressure	2 bar	2 bar	2 bar	2 bar
Nozzle type	Flat fan	Flat fan	Flat fan	Flat fan
Nozzle size	02F110	02F110	02F110	02F110
Application water volume/ha	600 L	600 L	600 L	600 L
Temperature of air - shade (°C)	17.4	23.0	21.5	22.1
Relative humidity (%)	95.9	66.8	51.9	60.1
Wind speed range (m/s)	0.4	0	0	0

Dew presence (Y/N)	N	N	N	N
Temperature of soil - 2-5 cm (°C)	-	-	-	-
Wetness of soil - 2-5 cm	Damp	Wet	Damp	Wet
Cloud cover (%)	100	100	100	100

### Untreated levels of pests/pathogens at application and through the assessment period

Common name	Scientific Name	EPPO Code	Infestation level pre-application	Infestation level at start of assessment period	Infestation level at end of assessment period
Melon and cotton aphid	<i>Aphis gossypii</i>	APHIGO	20.55 <sup>1</sup>	20.19 <sup>1</sup>	29.33 <sup>1</sup>
Peach-potato aphid	<i>Myzus persicae</i> <sup>2</sup>	MYZUPE	9.90 <sup>1</sup>	15.60 <sup>1</sup>	44.43 <sup>1</sup>

<sup>1</sup> Mean number of winged and wingless aphids per plant.

<sup>2</sup> Non target pest (natural infestation)

### Assessment details

Prior to the application of treatments, water-sensitive paper was used to demonstrate spray coverage, using water. Papers were attached to two representative *Hebe* plants using paper clips. Water was applied to the plants at 600 L / ha using the same equipment used for application of all the treatments in the trial. Spray coverage was then assessed by the deposition of droplets on the paper. For spray applications during the trial, the plants in each plot were removed from the cages for each respective treatment and replaced immediately after application.

*Aphis gossypii*, collected from a Hardy Nursery Stock nursery on cuttings of Hibiscus in April, was conditioned to *Hebe* cv. Purple Pixie by allowing them to breed on *Hebe* plants in insect proof cages in a polytunnel at ADAS Boxworth. By September, numbers of *A. gossypii* had built sufficiently to infest the trial plants. One marked stem on each of the trial plants was infested with approximately 20 adult, wingless aphids on 3 September, by placing a detached portion of *Hebe* on each of the marked stems.

The first assessment was carried out on 9 September, two days prior to the first treatment application so that plants could be ranked for numbers of aphids on the following day (one day before the first spray application). The ranking was done in order to account for the variability in aphid infestation success. Plants from each ranking group were then selected for each plot, in order to ensure that each plot was infested with similar numbers of aphids.

Treatments were applied over a 22-day period at time intervals recommended by each manufacturer. These varied from once, to twice at seven-day intervals and three times at five-day intervals.

Evaluation date	Evaluation Timing (DA)*		Crop Growth Stage (BBCH)	Evaluation type (efficacy, phytotox)	Assessment
	After conventional insecticides	After Bio-pesticides			
09/09/20	-2	-2	Inflorescence emerging	Efficacy and Phytotoxicity	Numbers of live wingless & winged & parasitised <i>A. gossypii</i> & <i>M. persicae</i> , aphid predators/parasitoids & phytotoxicity.
14/09/20	3	3	Inflorescence emerging	Efficacy and Phytotoxicity	Numbers of live wingless & winged & parasitised <i>A. gossypii</i> & <i>M. persicae</i> , aphid predators/parasitoids & phytotoxicity.
17/09/20	6	6	Flowering	Efficacy and Phytotoxicity	Numbers of live wingless & winged & parasitised <i>A. gossypii</i> & <i>M. persicae</i> , aphid predators/parasitoids & phytotoxicity. Numbers of <i>A. gossypii</i> and <i>M. persicae</i> infected with EPF.
24/09/20	13	13	Flowering	Efficacy and Phytotoxicity	Numbers of live wingless & winged & parasitised <i>A. gossypii</i> & <i>M. persicae</i> , aphid predators/parasitoids & phytotoxicity. Numbers of <i>A. gossypii</i> and <i>M. persicae</i> infected with EPF.
01/10/20	20	20	Flowering	Efficacy and Phytotoxicity	Numbers of live wingless & winged & parasitised <i>A. gossypii</i> & <i>M. persicae</i> , aphid predators/parasitoids & phytotoxicity. Numbers of <i>A. gossypii</i> and <i>M. persicae</i> infected with EPF.

\* DAT – days after first

## Statistical analysis

ANOVA (Genstat edition 18.2) was used to analyse the data. Numbers of wingless and winged aphids of each species per plant were combined for analysis. A log, base 10, transformation was applied to the data for additional clarification of statistical analysis using ANOVA with a significance level of  $P < 0.05$ . Differences between means were compared with Duncan's Multiple Range Test. Graphs are presented with back-transformed means in order to have relevance for growers. Abbott's formula was used to calculate percentage reduction in numbers of aphids compared with the untreated control.

## Results

### Spray coverage

Spray coverage was good in both the middle and upper canopy (Figures 1 and 2, Appendix) There was around 50% less spray reaching the middle canopy than the top canopy on one plant, but the spray coverage was similar in the middle and upper canopy of the second plant. A little spray reached the underside of a top leaf on one plant but there was no spray on the underside of the other leaves tested.

### Phytotoxicity

No phytotoxicity symptoms were observed on any assessment date.

### Efficacy

#### *Aphis gossypii*

On 9 September, two days before the first treatment application there were no statistically significant differences between treatments and the mean number of *A. gossypii* per plant was very similar in all treatments (**Table 1, Figure 1**). On 14 September, three days after the first treatment application, none of the treatments had significantly reduced the *A. gossypii* population compared with the untreated control (**Table 1, Figure 1**). *Aphis gossypii* were mostly found in the *Hebe* flower buds, on the stems and the underside of leaves during assessments (**Appendix, Figures 5 and 6**).

The second application of all bioprotectants was carried out one day before the next assessment on 17 September. On 17 September, AHDB9968, a bioprotectant, had reduced mean numbers of *A. gossypii* by 67.23% compared with the untreated control, which was six days after the first treatment application ( $P = 0.002$ ) (**Table 2**). On 24 September, after three applications of this product there was a reduction by 79.9% in mean numbers of *A. gossypii* compared with the untreated control ( $P = 0.002$ ). On 1 October, 10 days after the final application, this product achieved a 70.94% reduction in mean numbers of *A. gossypii* compared with the untreated control ( $P < 0.001$ ).

AHDB9921, a conventional insecticide did not reduce mean numbers of *A. gossypii* until the assessment on 1 October, 20 days after the first application and 13 days after the second application on 18 September. On 1 October, mean numbers of *A. gossypii* were reduced by 74.94% compared with the untreated control ( $P = < 0.001$ ) (**Table 2**). Prior to the assessment on 1 October, the *A. gossypii* infestation had been increasing, relative to the untreated control (**Table 2**).

The industry standard, flonicamid (Mainman) did not significantly reduce mean numbers of *A. gossypii* compared with the untreated control on any assessment date (**Tables 1 and 2**). In addition, mean numbers of *A. gossypii* were not significantly reduced by the bioprotectants AHDB9920, AHDB9919 and AHDB9918 compared with the untreated control or the industry standard, Mainman, on any assessment date.

Alate (winged) *A. gossypii* were found in all treatments throughout the trial. The percentage of alate aphids to apterous (wingless) aphids was low at the start of the experiment and had increased by the end of the experiment (**Table 3**).

**Table 1.** Transformed mean number of *Aphis gossypii* per plant. Transformation: Log base 10 (x+1) (back-transformed means are shown in brackets). Values not sharing the same letter are statistically significantly different (P<0.05).

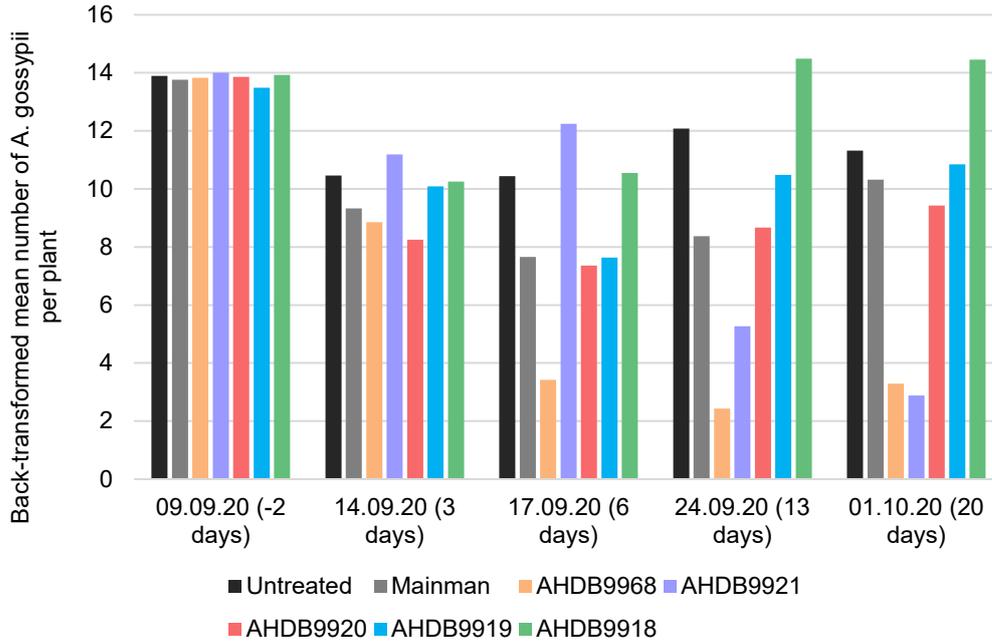
Date	09.09.20 (Day -2)	14.09.20 (Day 3)	17.09.20 (Day 6)	24.09.20 (Day 13)	01.10.20 (Day 20)
<b>Treatment</b>					
Untreated	1.173 a (13.89)	1.059 a (10.46)	1.0585 c (10.44)	1.1165 bc (12.08)	1.0907 b (11.32)
Mainman	1.169 a (13.76)	1.014 a (9.33)	0.9373 ab (7.66)	0.9718 bc (8.37)	1.0537 b (10.32)
AHDB9968	1.171 a (13.83)	0.994 a (8.86)	0.6456 a* (3.42)	0.5349 a* (2.43)	0.6325 a* (3.29)
AHDB9921	1.176 a (14.00)	1.086 a (11.19)	1.1218 bc (12.24)	0.7972 ab (5.27)	0.5883 a* (2.88)
AHDB9920	1.172 a (13.86)	0.966 a (8.25)	0.9222 bc (7.36)	0.9853 bc (8.67)	1.0183 b (9.43)
AHDB9919	1.161 a (13.49)	1.045 a (10.09)	0.9360 abc (7.63)	1.0601 bc (10.48)	1.0739 b (10.85)
AHDB9918	1.174 a (13.93)	1.051 a (10.25)	1.0625 bc (10.55)	1.1901 c (14.49)	1.1893 b (14.46)
F value	0.70	0.57	4.53	4.43	7.09
P value	0.648	0.752	0.002	0.002	<0.001
d.f.	36	36	36	36	36
s.e.d.	0.00839	0.0776	1.042	0.1490	0.1264
l.s.d.		0.1573	0.2113	0.3022	0.2563
	Not significantly different from untreated control (p>0.05)				
	*Significantly different from untreated control (p<0.05)				

**Table 2.** Abbott's formula values for percentage reduction in numbers of *Aphis gossypii* per plant compared with the untreated control.

Date	09.09.20 (Day -2)	14.09.20 (Day 3)	17.09.20 (Day 6)	24.09.20 (Day 13)	01.10.20 (Day 20)
<b>Treatment</b>					
Untreated					
Mainman	0.98	10.78	26.68	30.68	8.89
AHDB9968	0.49	15.23	67.23	79.90	70.94
AHDB9921	-0.74	-7.03	-17.19	56.37	74.94
AHDB9920	0.25	21.12	29.52	28.23	16.71
AHDB9919	2.92	3.48	26.93	13.19	4.13
AHDB9918	-0.25	2.00	-1.01	-20.00	-27.74

**Table 3.** Alate (winged) *A. gossypii* as a percentage of apterous (wingless) *A. gossypii* -2, 3, 6, 13 and 20 days after the first application of treatments.

Date	09.09.20 (Day -2)	14.09.20 (Day 3)	17.09.20 (Day 6)	24.09.20 (Day 13)	01.09.20 (Day 20)
<b>Treatment</b>					
Untreated	0.59	0.21	5.43	0.37	9.50
Mainman	0.28	0.93	6.67	0.31	12.46
AHDB9968	0.52	1.80	2.24	0.62	9.90
AHDB9921	0.37	0.00	1.00	2.27	10.35
AHDB9920	0.52	0.07	1.12	0.71	5.06
AHDB9919	0.04	0.09	0.79	0.70	7.48
AHDB9918	0.31	0.29	0.94	1.41	8.87



**Figure 1** Mean numbers of *A. gossypii* -2, 3, 6, 13 and 20 days after the first treatment application. Back-transformed data shown.

### ***Myzus persicae***

*Myzus persicae* were found naturally infesting the trial plants from the first assessment on 9 September. *Myzus persicae* were mostly found on the growing points of the *Hebe* plants. There were no differences in the mean numbers of *M. persicae* per plant between any of the treatments on 9 or 14 September (**Table 4**). On 17 September, six days after the first application and one day after the second application, AHDB9968 had reduced the population of *M. persicae* by 72.16% compared with the untreated control ( $P < 0.05$ , **Table 5**). On 24 September, 13 days after application, four products had reduced the population of *M. persicae* compared with the untreated control ( $P < 0.001$ ); AHDB9968, AHDB9921, AHDB9920 and AHDB9919. On this date AHDB9968 had reduced more of the *M. persicae* population than AHDB9920 and AHDB9919 (**Table 4**, **Figure 2**). On 1 October, 20 days after the first treatment application AHDB9968 and AHDB9921 had reduced the population of *M. persicae* when compared with the untreated control ( $P < 0.001$ ). Mainman, the industry standard did not reduce the population of *M. persicae* on any assessment date, although Abbott's formula showed that there had been a reduction in aphid numbers compared with the controls, this was not statistically significant (**Table 5**). The bioprotectant AHDB9918 did not reduce the population of *M. persicae* compared with the untreated control on any date (**Tables 4 and 5**).

Alate (winged) aphids were found in all treatments and the percentage of alate to apterous (wingless) *M. persicae* had increased in all treatments by the end of the trial (**Table 6**).

**Table 4.** Transformed mean number of *Myzus persicae* per plant. Transformation: Log base 10 (x+1) (back-transformed means are shown in brackets). Values not sharing the same letter are statistically significantly different (P<0.05).

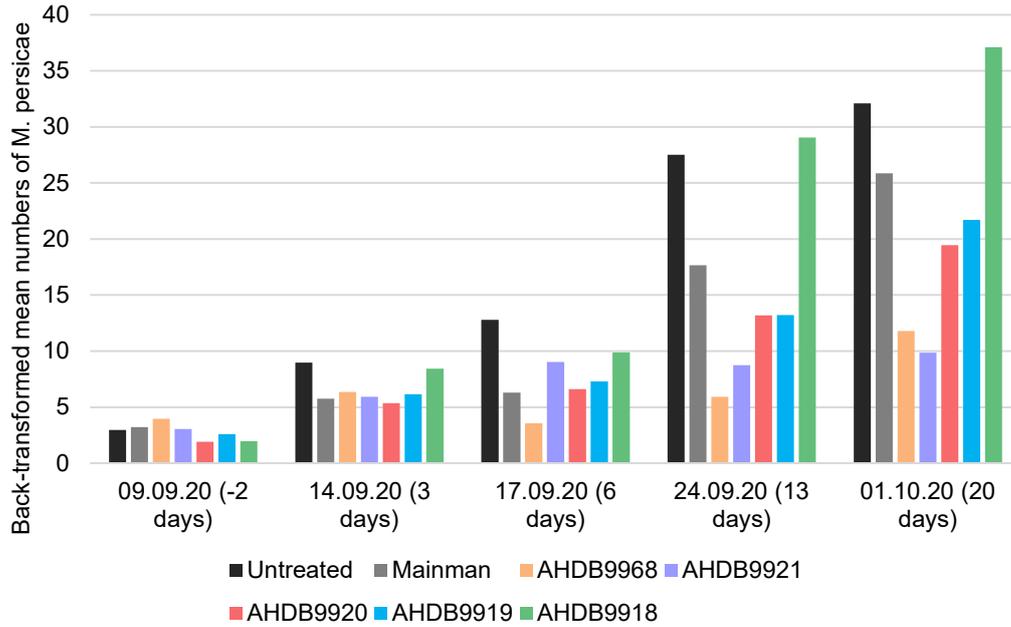
Date	09.09.20 (Day -2)	14.09.20 (Day 3)	17.09.20 (Day 6)	24.09.20 (Day 13)	01.10.20 (Day 20)
<b>Treatment</b>					
Untreated	0.5964 a (2.95)	0.9991 a (8.98)	1.1391 b (12.78)	1.455 c (27.51)	1.520 c (32.11)
Mainman	0.6267 a (3.23)	0.8298 a (5.76)	0.8635 ab (6.30)	1.271 bc (17.66)	1.429 c (25.85)
AHDB9968	0.6950 a (3.95)	0.8663 a (6.35)	0.6586 a* (3.56)	0.841 a* (5.93)	1.311 ab* (11.79)
AHDB9921	0.6080 a (3.06)	0.8408 a (5.93)	1.0018 b (9.04)	0.989 ab* (8.75)	1.107 a* (9.86)
AHDB9920	0.4656 a (1.92)	0.8038 a (5.37)	0.8806 ab (6.60)	1.152 b* (13.19)	1.311 bc (19.46)
AHDB9919	0.5544 a (2.58)	0.8543 a (6.15)	0.9181 ab (7.28)	1.153 b* (13.22)	1.356 bc (21.70)
AHDB9918	0.4739 a (1.98)	0.9752 a (8.44)	1.0371 b (9.89)	1.478 c (29.06)	1.581 c (37.11)
F value	0.91	0.96	2.86	5.75	5.57
P value	0.496	0.468	0.022	<0.001	<0.001
d.f.	36	36	36	36	36
s.e.d.	0.1226	0.1088	0.1281	0.1372	0.1214
l.s.d.		0.2206	0.2598	0.2783	0.2461
	Not significantly different from untreated control (p>0.05)				
	*Significantly different from untreated control (p<0.05)				

**Table 5.** Abbott's formula values for percentage reduction in numbers of *Myzus persicae* per plant compared with the untreated control.

Date	09.09.20 (Day -2)	14.09.20 (Day 3)	17.09.20 (Day 6)	24.09.20 (Day 13)	01.10.20 (Day 20)
<b>Treatment</b>					
Untreated					
Mainman	-9.68	35.88	50.66	35.79	19.49
AHDB9968	-34.13	29.28	72.16	78.43	63.27
AHDB9921	-3.63	33.95	29.23	68.19	69.28
AHDB9920	34.83	40.25	48.37	52.05	39.39
AHDB9919	12.34	31.51	43.00	51.93	32.43
AHDB9918	32.91	5.95	22.57	-5.64	-15.55

**Table 6.** Alate (winged) *M. persicae* as a percentage of apterous (wingless) *M. persicae*.

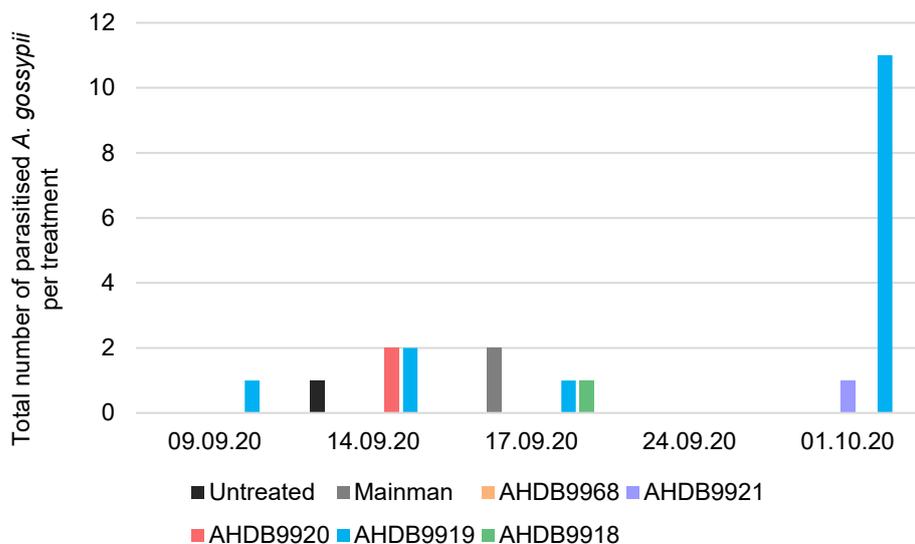
Date	09.09.20 (Day -2)	14.09.20 (Day 3)	17.09.20 (Day 6)	24.09.20 (Day 13)	01.09.20 (Day 20)
<b>Treatment</b>					
Untreated	0.28	4.37	0.84	2.17	8.45
Mainman	0.00	4.20	2.78	4.82	7.43
AHDB9968	0.00	7.25	4.99	2.15	5.97
AHDB9921	0.27	2.47	2.42	5.84	8.94
AHDB9920	0.53	2.74	1.95	3.10	7.78
AHDB9919	0.00	2.34	1.11	5.82	9.11
AHDB9918	1.14	2.41	1.97	2.50	11.45



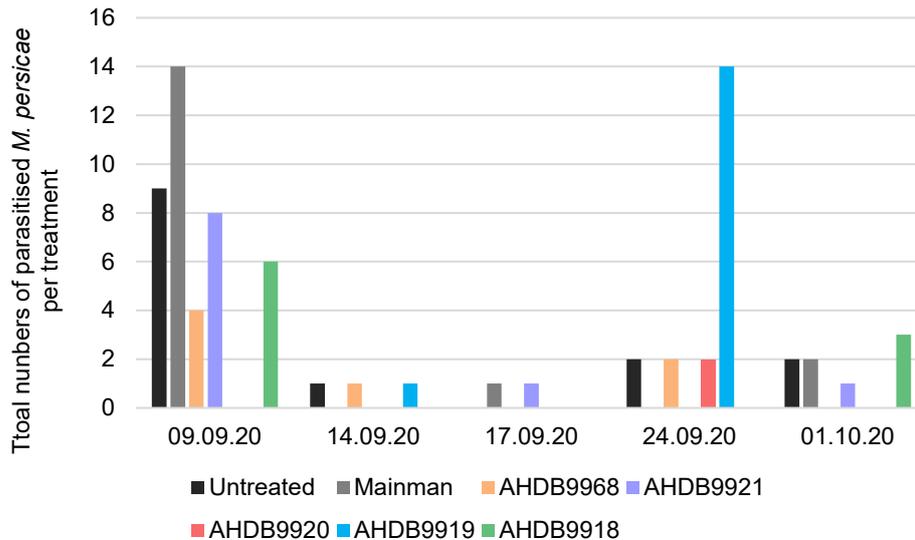
**Figure 2** Mean number of *M. persicae* per plant -2, 3, 6, 13 and 20 days after the first treatment application. Back-transformed data shown.

### Parasitoids and predators

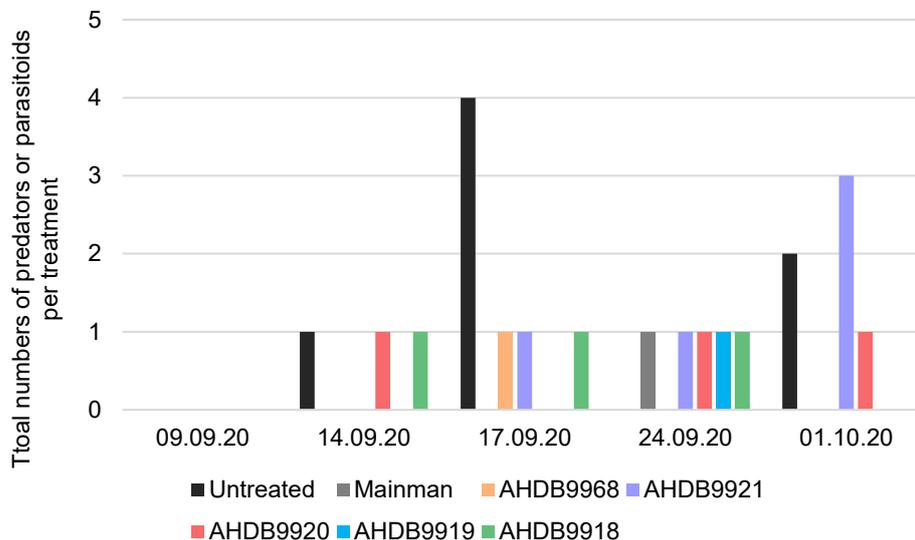
Parasitoids and parasitised aphids were recorded in the trial but numbers were too low for data analysis (**Figures 3, 4 and 5**). Parasitoid adults and mummies (parasitised aphids) recorded resembled *Praon volucre* and *Aphidius colemani* but were not identified to species. Predators included hoverfly adults, larvae and spiders. Some dead parasitoids were found stuck to plant leaves, which may have been killed by the treatment application. One dead parasitoid was found in single plots of the untreated control, AHDB9921 and AHDB9918 treatments.



**Figure 3** Total number of parasitised *A. gossypii* recorded per treatment on each assessment date.



**Figure 4** Total number of parasitised *M. persicae* per treatment on each assessment date.



**Figure 5** Total number of adult parasitoids and aphid predators recorded per treatment on each assessment date.

## Entomopathogenic Fungus

A naturally occurring entomopathogenic fungus (EPF), likely to have been *Pandora neoaphidis*, (Prince, personal communication, 2020) was noticed in the trial on 14 September and the numbers of aphids infected by this EPF were recorded on the following assessments. On 17 September there were significantly more *A. gossypii* killed by EPF in the untreated control than in all the other treatments, except for AHDB9921 (Table 7). On 24 September the highest mean number of *A. gossypii* killed by EPF was in the untreated control, which was significantly greater than in three of the other treatments. By 1 October there were significantly more *A. gossypii* killed by EPF in the untreated control than in any of the other treatments. However, in addition to numbers of infected aphids, there were also more healthy aphids in the untreated control plots than in the treated plots. Therefore the percentage of infected aphids in each plot were compared and there were no significant differences between treatments in the percentage of aphids killed by EPF (out of the total *A. gossypii* i.e. live plus infected) on any assessment date (Table 7).

**Table 7.** Mean numbers of *Aphis gossypii* per plant killed by EPF, untransformed data and percentage aphids killed by EPF of total aphids. Values with different letters are significantly different (P <0.05).

Date	17.09.20 (Day 6)		24.09.20 (Day 13)		01.10.20 (Day 20)	
	Mean per plant	% of total <i>A. gossypii</i>	Mean per plant	% of total <i>A. gossypii</i>	Mean per plant	% of total <i>A. gossypii</i>
Untreated	0.9524 b	7.1	1.8571 b	13.0	8.190 b	30.6
Mainman	0.1905 a*	3.3	0.5238 a*	4.2	2.714 a*	15.5
AHDB9968	0.2143 a*	8.2	0.4048 a*	6.3	1.190 a*	5.8
AHDB9921	0.4286 ab	3.5	0.8095 ab	7.7	0.619 a*	9.6
AHDB9920	0.0714 a*	2.4	0.1905 a*	5.5	1.143 a*	11.7
AHDB9919	0.1667 a*	1.4	0.9762 ab	4.3	2.429 a*	11.6
AHDB9918	0.0714 a*	2.3	1.0000 ab	8.6	4.310 a*	17.0
F value	2.3	0.85	1.94	0.70	5.06	1.63
P value	0.055	0.539	0.100	0.653	<0.001	0.168
d.f.	36	36	36	36	36	36
s.e.d.	0.2908	3.97	0.554	5.24	1.652	8.87
l.s.d.	0.5897	8.05	1.123	10.62	3.350	17.99
	Not significantly different from untreated control (p>0.05)					
	*Significantly different from untreated control (p<0.05)					

The same EPF also affected *M. persicae*, with higher mean numbers of *M. persicae* killed by the EPF than *A. gossypii*. There were no statistically significant differences between treatments on the first assessment, 17 September (**Table 8**). Significantly more *M. persicae* were killed by EPF in the untreated control than three of the treatments on 24 September and significantly more were killed in the untreated control compared with four of the treatments on 1 October. However, as for *A. gossypii*, when the percentage of total aphids (live plus infected) *M. persicae* per plot were compared, there were no significant differences between treatments in the percentage of aphids killed by EPF (**Table 8**).

**Table 8.** Mean number of *Myzus persicae* per plant killed by EPF, untransformed data and percentage aphids killed by EPF of total aphids. Values with different letters are significantly different (P<0.05).

Date	17.09.20 (Day 6)		24.09.20 (Day 13)		01.10.20 (Day 20)	
	Mean per plant	% of total <i>M. persicae</i>	Mean per plant	% of total <i>M. persicae</i>	Mean per plant	% of total <i>M. persicae</i>
Untreated	1.476 a	3.89	5.929 b	10.5	11.786 c	20.0
Mainman	0.714 a	5.08	1.119 a*	3.2	5.286 ab*	10.5
AHDB9968	0.976 a	11.34	1.143 a*	5.7	1.667 a*	5.7
AHDB9921	1.571 a	7.19	2.976 ab	13.6	3.024 ab*	11.8
AHDB9920	0.738 a	3.51	1.405 a*	3.5	4.738 ab*	10.3
AHDB9919	1.095 a	4.7	3.333 ab	6.5	6.190 abc	11.2
AHDB9918	1.762 a	5.08	3.667 ab	7.1	8.690 bc	15.2
F	0.45	1.24	1.80	1.81	3.36	1.87
P	0.842	0.310	0.126	0.125	0.010	0.113
d.f.	36	36	36	36	36	36
s.e.d.	0.880	3.425	1.843	3.93	2.643	4.62
l.s.d.	1.785	6.947	3.737	7.98	5.360	9.36
	Not significantly different from untreated control (p>0.05)					
	*Significantly different from untreated control (p<0.05)					

## Discussion

The target aphid species in this trial was *A. gossypii*, which was released onto the plants before products were applied. However, a natural infestation of *M. persicae* was detected on the plants from the first assessment date. The trial plants had been sourced from a commercial nursery and were kept in insect-proof cages in a polytunnel at ADAS Boxworth prior to being placed in smaller cages (BugDorms®) when used in the trial. It is possible that small numbers of *M. persicae* were present on the plants when they arrived from the nursery, or winged *M. persicae* might have infested the plants at ADAS Boxworth, e.g. when the cages were opened for additional irrigation or for selecting plants for the trial. However, the natural infestation with *M. persicae* allowed information to be gained on efficacy of the products against this species in addition to *A. gossypii*. None of the treatments eliminated either *A. gossypii* or *M. persicae* within the trial period, from a starting mean of 13 -14 *A. gossypii* and 1 – 4 *M. persicae* per plant.

The botanical bioprotectant AHDB9968, applied three times at 5-day intervals performed well against both *A. gossypii* and *M. persicae* with a maximum reduction of 79.90% *A. gossypii* and 78.43% *M. persicae* compared with untreated controls. Although this product did not give a quick knockdown of aphids three days after the first application, it significantly reduced populations of both species from six days after the first application. Ten days after the final application, this treatment had still significantly reduced the *A. gossypii* and *M. persicae* populations compared with the untreated control. However, on the final assessment date, the mean number of aphids per plant had started to rise, indicating that another product treatment might be needed at this stage to prevent the population building up again. These results differ from an experiment on *Hebe* in 2016 when AHDB9968 was applied at the same rate and timing but there was no significant reduction when compared with the water control (Pope *et al.*, 2017). However, Pope *et al.* (2016) and Pope *et al.* (2015) found a significant reduction in numbers of *A. gossypii* on *Hebe* and *M. persicae* on pansy respectively when AHDB9968 was applied at 1.8 L / ha in 600 L water four times at weekly intervals. AHDB9968 was ineffective when tested against *A. gossypii* on strawberry and *M. persicae* on Brussels sprout (Fountain *et al.*, 2019; Collier & Jukes, 2019).

The conventional insecticide AHDB9921, applied twice at 7-day intervals was not effective against *A. gossypii* until after the second application, when a significant reduction of 74.94% compared with the untreated control was achieved 20 days after the first application and 13 days after the second application, indicating that the product could have a delayed effect. This insecticide was effective against *M. persicae* from 13 days after the first application, suggesting that this product is more effective against *M. persicae* than *A. gossypii*.

Mainman, the industry standard conventional insecticide was ineffective against *A. gossypii* and *M. persicae* in this trial. Although it is possible that these particular aphid populations might have developed resistance to flonicamid, it is unlikely, as IRAC Sucking pest WG (2019) state that only one report of *A. gossypii* resistance to flonicamid has been reported, in Korea on pepper, and IRAG UK (2020) state that ongoing resistance screening has shown that there is no evidence of resistance to flonicamid in *M. persicae* in the UK. Gore *et al.* (2013) found that *A. gossypii* was not resistant to flonicamid when tested, but reported that there is likely to be a high selection pressure for *A. gossypii* to develop resistance to an insecticide with a high level of efficacy at a low rate given the high reproductive capacity of *A. gossypii*.

Mainman was applied only once in this trial, although three applications per year are permitted under EAMU 0045/2013, an interval of 21 days must be left between applications. Mainman is often used as part of an IPM programme rather than being used as the sole means of control. Mainman reduced numbers of *A. gossypii* and *M. persicae* relative to the untreated control but the difference was not significant. This result with *A. gossypii* differed from the findings of Pope *et al.* (2016 & 2017) where Mainman gave significant reductions in numbers of *A. gossypii* on *Hebe* compared with the water control. In these trials, Mainman was effective six days after the second application seven days after the first in 2014 and six days after a single application in 2016. However, the experiments conducted by Pope *et al.* were carried out in July and August when it is likely that there was more soft growth on the *Hebe* plants. As *Hebe* leaves have a thick waxy cuticle, soft plant tissue may have been easier for Mainman to penetrate, compared with the older growth in this experiment. Flonicamid is a systemic product, and therefore should not have been affected by any lack of spray deposition on leaf undersides. Spray deposition on the *Hebe* plants was similar in the experiments carried out by Pope *et al.* and in this experiment.

The botanical bioprotectants AHDB9920 and AHDB9919 were effective against *M. persicae* after three applications at 5-day intervals, however performance against *A. gossypii* was comparable to Mainman, the industry standard and aphid numbers did not significantly differ from the control. These contact-acting products might be useful against *M. persicae*, alongside released biocontrols, and used to rotate modes of action to prevent resistance developing. Contact-acting products tend to be less useful against *A. gossypii*, which is protected by waxy secretions and the aphids were found in the flower buds, which provided protection from spray deposits.

The contact-acting botanical bioprotectant AHDB9918 was ineffective against both *A. gossypii* and *M. persicae*.

Low numbers of aphid predators, parasitoids and parasitised aphids were found in plots of all the treatments despite the trial plants being enclosed in insect-proof cages (Bugdorms®) to reduce immigration by any other pests or natural enemies. The aphid predators and parasitoids could have gained entry to the cages when they were opened for assessments or when the plants were taken out of the cages for spray application. As these biological control agents were found in plots of all treatments, this indicates that the treatments should be compatible with these biocontrols if used in an IPM programme. Mainman is reported to be safe (<25% mortality) to lacewing larvae and slightly harmful (25-50% mortality) to *Aphidius* spp. and hoverfly larvae (Biobest 2020; Koppert Biological Systems, 2020). AHDB9968 is reported to be safe to *Aphidius* spp., lacewing larvae and slightly harmful to hoverfly larvae. AHDB9920 and AHDB9918 are reported to be harmful (>75% mortality) to *Aphidius* spp. and harmful or moderately harmful (50-75% mortality) to lacewing larvae, although the persistence of these products is likely to be short. Three dead parasitoids were found, however one of these was found in an untreated control plot and therefore they may not have been killed by the treatments applied. Since numbers of aphid predators, parasitoids and parasitised aphids were too low for statistical analysis the presence of parasitoids and predators in this trial will not have had a significant effect on product efficacy.

Mean numbers of *A. gossypii* in the untreated controls dropped slightly between 9 and 14 September, which might have been due to infection with the EPF, which was discovered on 14 September. The temperature rose between these dates, therefore reproductive rate of *A. gossypii* would not be expected to fall due to temperature. Average relative humidity rose by 20% during the trial, providing ideal conditions for the EPF, which prefers humid environments. The pots of *Hebe* were stood on capillary

matting, which needed frequent watering during the hot conditions in the polytunnel during the trial. The 'BugDorm' cages are likely to have helped to maintain a high relative humidity around the plants. Significantly higher numbers of aphids were killed by EPF in the untreated control. However, the percentage of *A. gossypii* or *M. persicae* infected with EPF out of the total numbers of *A. gossypii* or *M. persicae* did not significantly differ between treatments due to the variability in natural infection. This suggests that the higher mean number of infected aphids in the untreated control was related to there being a greater total number of aphids in the untreated control and therefore more aphids available to infect. Since the proportion of aphids killed by the EPF was not significantly higher in the untreated controls than in other treatments, the presence of the EPF should not have prevented the treatments from giving significant reductions in numbers of live aphids in comparison to the untreated control.

The percentage of alate to apterous *A. gossypii* and *M. persicae* increased in all treatments during the trial, which is likely to be a response to crowding, temperature, day length and nutritional factors (Ebert & Cartwright, 1997).

## Conclusions

- The botanical bioprotectant AHDB9968 applied three times at 5-day intervals provided control of *A. gossypii* and *M. persicae* from six days after the first application.
- The conventional insecticide AHDB9921 applied twice at 7-day intervals provided a significant reduction in the number of *A. gossypii* compared with the untreated control 13 days after the second application and 20 days after the first application. This insecticide also provided a significant reduction in the number of *M. persicae* compared with the untreated control from 13 days after the first application.
- The botanical bioprotectants AHDB9920 and AHDB9919 applied three times at 5-day intervals were not effective against *A. gossypii* but were effective against *M. persicae* on one assessment date, after three applications at 5-day intervals.
- The industry standard flonicamid (Mainman) did not reduce numbers of *A. gossypii* or *M. persicae* compared with the untreated control in this experiment.
- The bioprotectant AHDB9918 was ineffective against both *A. gossypii* and *M. persicae*.
- The EPF, likely to be *Pandora neoaphidis*, killed more aphids in the untreated control than in some of the other treatments. However, as there were also more healthy aphids in the control plots in addition to more infected aphids, the proportion of EPF-infected aphids was not higher than in the treated plots and therefore should not have impacted on the reduction in numbers of live aphids by the tested treatments.
- No treatments caused phytotoxic effects on the tested plant species, *Hebe* var. Purple Pixie

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## **Acknowledgements**

With thanks to the AHDB for funding and supporting this project and for the financial and in-kind contributions from ICL, The Bransford Webbs Plant Company and the crop protection manufactures, and distributors involved with the SCEPTREplus programme as listed below:

Agrii, Alpha Biocontrol Ltd, Andermatt, Arysta Lifescience, BASF, Bayer, Belchim, Bionema Limited, Certis Europe, Dow, DuPont, Eden Research, Fargro Limited, FMC, Gowan, Interfarm, Lallemand Plant Care, Novozymes, Oro Agri, Russell IPM, Sumitomo Chemicals, Syngenta, UPL.

## Appendix

a. Crop diary – events related to growing crop

Crop	Cultivar	Potting up date	Pots per cage
<i>Hebe</i>	Purple pixie	15/04/20	6

Date	Event
6/04/20	<i>Hebe</i> plugs received at ADAS Boxworth.
15/04/20	<i>Hebe</i> plugs potted and placed into insect proof tents in a poly tunnel.
17/04/20	<i>Aphis gossypii</i> acquired from local nursery on hibiscus cuttings and transferred to <i>Hebe</i> , kept in a BugDorm in a poly tunnel.
24/06/20	<i>Aphis gossypii</i> conditioned and successfully reproducing on <i>Hebe</i> .
03/09/20	Plants trimmed to 15 cm.
03/09/20	Infestation with 20 aphids per plant.
04/09/20	Biocontrol released for two-spotted spider mite ( <i>Phytoseiulus persimilis</i> )

Biological control agents applied for other pests

Date	Product	Rate per cage	Pest
04/09/20	<i>Phytoseiulus persimilis</i>	20	<i>Tetranychus urticae</i>

Details of irrigation regime

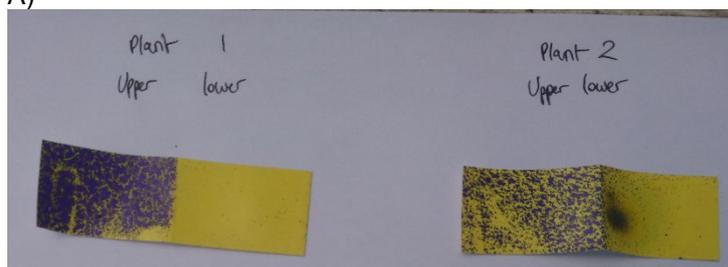
Plants were irrigated by hand onto capillary matting lining the bottom of each BugDorm. The matting was kept damp throughout the trial.

b. Trial diary

	Day	
03/09/2020		Infestation complete. Approximately 20 adult aphids per plant
09/09/2020	0	Pre-randomisation assessment completed
10/09/2020	1	Plants ranked and put into blocks with the same average number of aphids per plot. Plot number / treatment assigned
11/09/2020	2	First spray completed.
14/09/2020	5	Assessment 2 completed
16/09/2020	7	Second spray complete. T3,5,6, & 7 applied
17/09/2020	8	Assessment 3 completed
18/09/2020	9	3rd spray completed. T4 applied.
21/09/2020	12	Final (4 <sup>th</sup> ) spray completed. T3, 5, 6, & 7 applied.
24/09/2020	15	Assessment 4 partially completed, except for four plots, as daylight ran out.
25/09/2020	16	Assessment 4 four remaining plots assessed at sunrise, 7:00 am.
01/10/2020	22	Assessment 5 completed.

c. Photographs

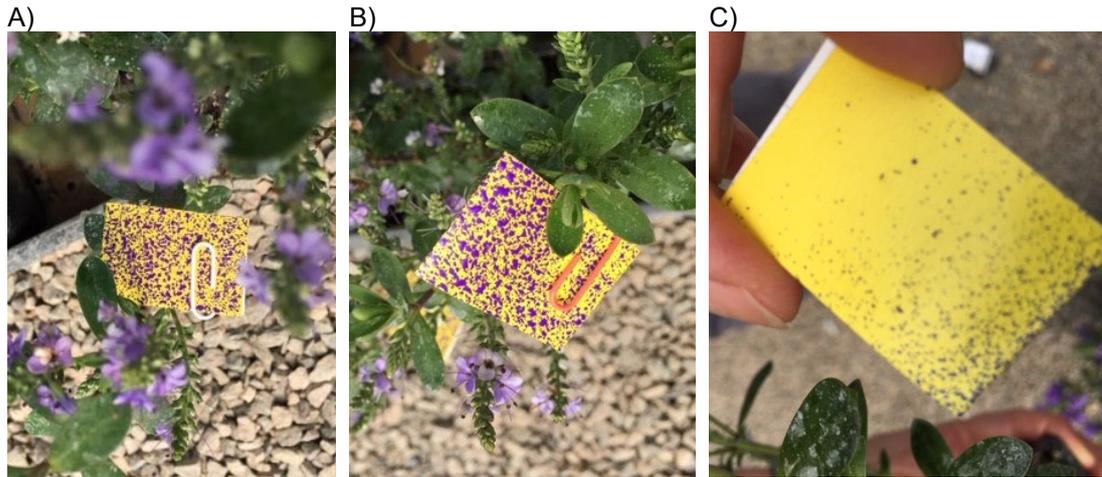
A)



B)



**Figure 1.** A) *Hebe* top canopy plant 1 (left) and plant 2 (right), upper leaf surface on the left of the paper and lower leaf surface on the right of the paper. B) *Hebe* middle canopy plant 1 (left) and plant 2 (right), upper leaf surface on the left of the paper and lower leaf surface on the right of the paper.



**Figure 2.** A) *Hebe* test spray middle canopy. B) *Hebe* test spray top canopy. C) *Hebe* test spray underside of top canopy.



**Figure 3.** Caged trial set up



**Figure 4.** One plot of *Hebe* plants in a BugDorm. Infested stems marked with red wool.

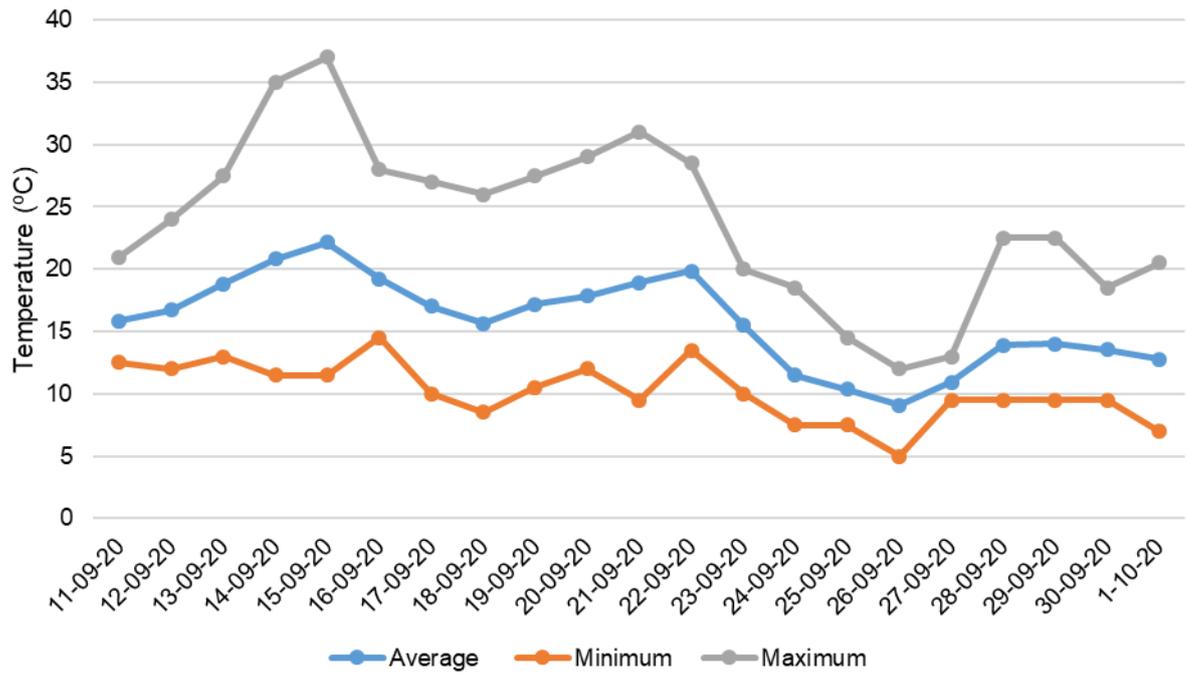


**Figure 5.** *Aphis gossypii* on a *Hebe* stem.

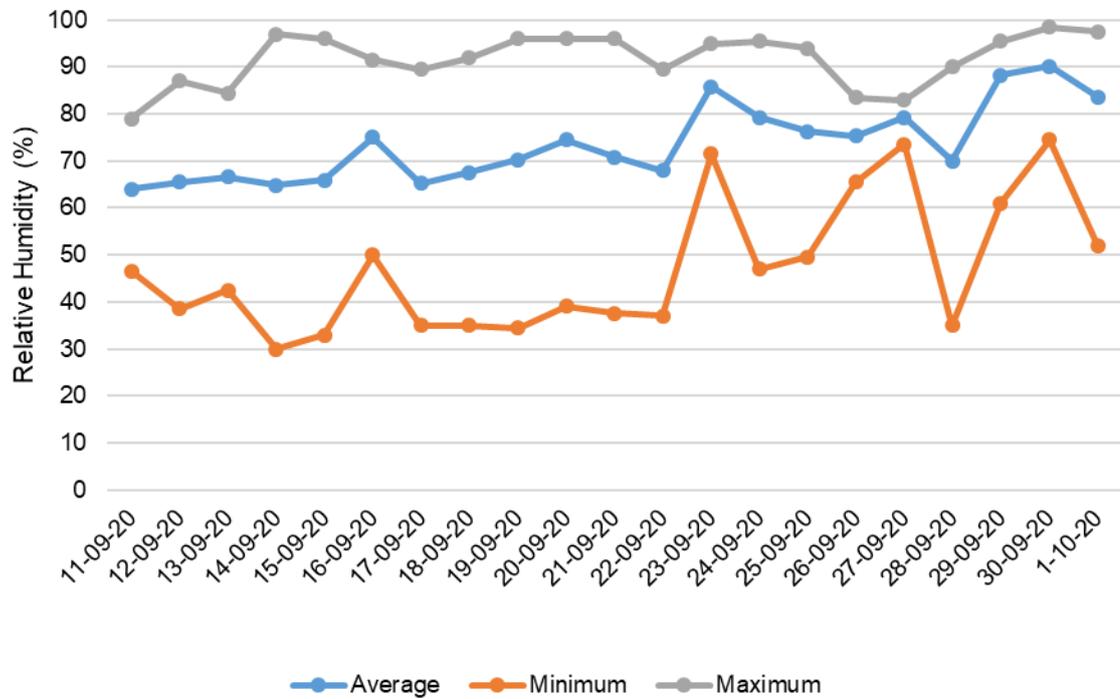


**Figure 6.** *Aphis gossypii* in *Hebe* buds.

d. Climatological data during study period



**Figure 2** Daily minimum, maximum and average temperature inside a central plot 'BugDorm' from 11 September until 1 October.



**Figure 3** Daily maximum, minimum and average relative humidity inside a central plot 'BugDorm' from 11 September until 1 October.

e. Raw data from assessments

09.9.20						
Plot	Treatment	Wingless <i>A.</i> <i>gossypii</i>	Winged <i>A.</i> <i>gossypii</i>	Wingless <i>M.</i> <i>persicae</i>	Winged <i>M.</i> <i>persicae</i>	Phytotoxicity <sup>1</sup>
101	5	2.83	0.00	7.00	0.00	1
102	1	2.83	0.00	5.33	0.00	1
103	4	2.83	0.00	7.33	0.00	1
104	7	2.83	0.00	14.00	0.00	1
105	6	2.83	0.00	13.50	0.00	1
106	2	2.67	0.00	2.50	0.00	1
107	3	2.50	0.00	6.67	0.00	1
201	2	4.83	0.00	8.83	0.00	1
202	5	5.00	0.00	7.00	0.00	1
203	6	4.67	0.00	2.50	0.00	1
204	4	5.33	0.00	0.67	0.00	1
205	3	5.67	0.17	3.67	0.00	1
206	1	5.33	0.17	2.83	0.00	1
207	7	5.33	0.00	8.33	0.00	1
301	3	10.33	0.00	9.33	0.00	1
302	1	10.50	0.00	13.00	0.00	1
303	4	10.50	0.17	9.00	0.00	1
304	7	10.50	0.00	5.00	0.00	1
305	2	11.83	0.00	8.50	0.00	1
306	5	10.50	0.00	2.83	0.00	1
307	6	10.50	0.00	10.33	0.00	1
401	7	15.17	0.17	5.67	0.00	1
402	5	15.50	0.00	2.50	0.00	1
403	3	15.33	0.00	9.83	0.00	1
404	6	15.17	0.00	1.67	0.00	1
405	1	16.00	0.00	22.50	0.00	1
406	4	15.33	0.00	8.83	0.00	1
407	2	15.17	0.17	12.00	0.00	1
501	3	20.50	0.00	15.83	0.00	1
502	2	20.50	0.00	9.33	0.00	1
503	6	20.50	0.00	17.83	0.00	1
504	1	20.50	0.00	3.67	0.00	1
505	4	21.67	0.17	15.00	0.00	1
506	5	20.50	0.17	10.50	0.00	1
507	7	20.67	0.00	0.33	0.00	1
601	1	29.67	0.17	13.50	0.00	1
602	3	31.00	0.00	12.33	0.00	1
603	5	30.83	0.33	4.33	0.17	1
604	2	30.83	0.00	4.83	0.00	1
605	7	31.00	0.33	1.67	0.00	1
606	6	30.67	0.00	5.50	0.00	1
607	4	31.00	0.00	8.50	0.17	1
701	3	61.67	0.50	6.00	0.00	1
702	1	58.33	0.33	8.33	0.17	1
703	6	57.17	0.17	3.33	0.00	1
704	2	56.33	0.50	11.83	0.00	1
705	5	56.50	1.00	2.17	0.00	1
706	4	56.17	0.17	9.83	0.00	1
707	7	57.83	0.00	3.83	0.33	1

<sup>1</sup> Phytotoxicity on a scale from 1 – 10 where 1 is no damage, comparable with control and 10 is complete crop kill

09.09.20				
Plot	Treatment	Parasitised <i>A. gossypii</i>	Parasitised <i>M. persicae</i>	Aphid predators
101	5	0.00	0.00	0.00
102	1	0.00	0.33	0.00
103	4	0.00	0.17	0.00
104	7	0.00	0.50	0.00
105	6	0.00	0.00	0.00
106	2	0.00	0.00	0.00
107	3	0.00	0.17	0.00
201	2	0.00	0.00	0.00
202	5	0.00	0.00	0.00
203	6	0.00	0.00	0.00
204	4	0.00	0.00	0.00
205	3	0.00	0.00	0.00
206	1	0.00	0.00	0.00
207	7	0.00	0.33	0.00
301	3	0.00	0.00	0.00
302	1	0.00	0.67	0.00
303	4	0.00	1.00	0.00
304	7	0.00	0.17	0.00
305	2	0.00	2.33	0.00
306	5	0.00	0.00	0.00
307	6	0.00	0.00	0.00
401	7	0.00	0.00	0.00
402	5	0.00	0.00	0.00
403	3	0.00	0.00	0.00
404	6	0.00	0.00	0.00
405	1	0.00	0.33	0.00
406	4	0.00	0.00	0.00
407	2	0.00	0.00	0.00
501	3	0.00	0.00	0.00
502	2	0.00	0.00	0.00
503	6	0.00	0.00	0.00
504	1	0.00	0.17	0.00
505	4	0.00	0.00	0.00
506	5	0.00	0.00	0.00
507	7	0.00	0.00	0.00
601	1	0.00	0.00	0.00
602	3	0.00	0.50	0.00
603	5	0.00	0.00	0.00
604	2	0.00	0.00	0.00
605	7	0.00	0.00	0.00
606	6	0.17	0.00	0.00
607	4	0.00	0.00	0.00
701	3	0.00	0.00	0.00
702	1	0.00	0.00	0.00
703	6	0.00	0.00	0.00
704	2	0.00	0.00	0.00
705	5	0.00	0.00	0.00
706	4	0.00	0.17	0.00
707	7	0.00	0.00	0.00

14.9.20						
Plot	Treatment	Wingless <i>A.</i> <i>gossypii</i>	Winged <i>A.</i> <i>gossypii</i>	Wingless <i>M.</i> <i>persicae</i>	Winged <i>M.</i> <i>persicae</i>	Phytotoxicity <sup>1</sup>
101	5	2.50	0.00	12.17	0.17	1
102	1	3.33	0.00	6.50	0.33	1
103	4	5.00	0.00	7.33	0.33	1
104	7	2.17	0.00	24.33	0.83	1
105	6	5.67	0.00	22.00	0.33	1
106	2	3.50	0.00	4.50	0.17	1
107	3	2.00	0.00	7.83	1.17	1
201	2	3.33	0.00	30.67	1.33	1
202	5	2.50	0.00	19.33	1.00	1
203	6	2.67	0.00	2.33	0.00	1
204	4	7.33	0.00	8.00	0.17	1
205	3	5.33	0.00	7.50	0.17	1
206	1	4.50	0.00	10.83	1.50	1
207	7	3.33	0.00	17.00	0.00	1
301	3	12.33	0.00	6.50	0.00	1
302	1	3.83	0.00	30.67	0.67	1
303	4	7.00	0.00	9.83	0.33	1
304	7	12.00	0.00	25.67	0.17	1
305	2	7.83	0.00	5.17	0.50	1
306	5	2.50	0.00	12.50	0.17	1
307	6	8.17	0.00	12.50	0.33	1
401	7	17.50	0.00	11.00	0.50	1
402	5	23.83	0.00	5.50	0.00	1
403	3	10.83	0.00	10.17	0.00	1
404	6	15.33	0.00	7.67	0.00	1
405	1	27.00	0.00	15.33	0.50	1
406	4	11.33	0.00	9.67	0.50	1
407	2	16.67	0.17	9.67	0.17	1
501	3	10.00	1.00	12.17	1.67	1
502	2	11.83	0.00	25.33	3.17	1
503	6	14.50	0.00	26.17	2.67	1
504	1	23.33	0.17	17.83	0.00	1
505	4	17.83	0.00	26.33	0.33	1
506	5	17.33	0.00	18.83	1.33	1
507	7	15.83	0.00	16.00	0.50	1
601	1	22.67	0.17	14.17	1.00	1
602	3	17.17	0.50	19.50	3.50	1
603	5	33.17	0.17	6.33	0.33	1
604	2	22.17	0.17	8.50	0.00	1
605	7	24.33	0.50	10.67	0.00	1
606	6	28.50	0.00	10.67	0.33	1
607	4	25.17	0.00	11.17	0.17	1
701	3	48.83	0.33	3.67	0.33	1
702	1	56.33	0.00	9.67	0.17	1
703	6	52.67	0.33	8.17	0.00	1
704	2	43.17	2.17	14.83	0.00	1
705	5	29.33	0.00	1.83	0.00	1
706	4	45.17	0.00	11.50	0.00	1
707	7	52.50	0.00	5.67	0.33	1

<sup>1</sup> Phytotoxicity on a scale from 1 – 10 where 1 is no damage, comparable with control and 10 is complete crop kill

14.09.20				
Plot	Treatment	Parasitised <i>A. gossypii</i>	Parasitised <i>M.</i> <i>persicae</i>	Aphid predators
101	5	0.17	0.00	0.17
102	1	0.00	0.00	0.00
103	4	0.00	0.00	0.00
104	7	0.00	0.00	0.00
105	6	0.17	0.00	0.00
106	2	0.00	0.00	0.00
107	3	0.00	0.00	0.00
201	2	0.00	0.00	0.00
202	5	0.00	0.00	0.00
203	6	0.00	0.00	0.00
204	4	0.00	0.00	0.00
205	3	0.00	0.00	0.00
206	1	0.00	0.00	0.00
207	7	0.00	0.00	0.00
301	3	0.00	0.00	0.00
302	1	0.17	0.17	0.00
303	4	0.00	0.00	0.00
304	7	0.00	0.00	0.00
305	2	0.00	0.00	0.00
306	5	0.00	0.00	0.00
307	6	0.17	0.00	0.00
401	7	0.00	0.00	0.17
402	5	0.17	0.00	0.00
403	3	0.00	0.00	0.00
404	6	0.00	0.00	0.00
405	1	0.00	0.00	0.17
406	4	0.00	0.00	0.00
407	2	0.00	0.00	0.00
501	3	0.00	0.17	0.00
502	2	0.00	0.00	0.00
503	6	0.00	0.17	0.00
504	1	0.00	0.00	0.00
505	4	0.00	0.00	0.00
506	5	0.00	0.00	0.00
507	7	0.00	0.00	0.00
601	1	0.00	0.00	0.00
602	3	0.00	0.00	0.00
603	5	0.00	0.00	0.00
604	2	0.00	0.00	0.00
605	7	0.00	0.00	0.00
606	6	0.00	0.00	0.00
607	4	0.00	0.00	0.00
701	3	0.00	0.00	0.00
702	1	0.00	0.00	0.00
703	6	0.00	0.00	0.00
704	2	0.00	0.00	0.00
705	5	0.00	0.00	0.00
706	4	0.00	0.00	0.00
707	7	0.00	0.00	0.00

17.9.20						
Plot	Treatment	Wingless <i>A.</i> <i>gossypii</i>	Winged <i>A.</i> <i>gossypii</i>	Wingless <i>M.</i> <i>persicae</i>	Winged <i>M.</i> <i>persicae</i>	Phytotoxicity <sup>1</sup>
101	5	3.50	0.00	26.67	0.83	1
102	1	4.67	0.00	13.00	0.17	1
103	4	10.17	0.17	16.50	1.00	1
104	7	2.67	0.00	28.17	1.33	1
105	6	6.33	0.00	32.33	0.17	1
106	2	7.00	0.00	9.83	0.00	1
107	3	0.83	0.00	6.33	0.67	1
201	2	2.50	0.00	20.50	0.50	1
202	5	1.83	0.00	21.83	0.33	1
203	6	3.00	0.00	5.67	0.00	1
204	4	8.50	0.00	11.83	0.00	1
205	3	3.17	0.00	3.50	0.17	1
206	1	7.33	0.00	24.33	0.33	1
207	7	3.83	0.00	29.83	0.00	1
301	3	6.17	0.00	3.33	0.00	1
302	1	4.50	0.00	71.83	0.00	1
303	4	13.17	0.33	19.17	0.50	1
304	7	21.50	0.17	16.33	0.67	1
305	2	7.67	0.00	9.17	0.33	1
306	5	4.17	0.00	13.83	1.17	1
307	6	12.33	0.00	17.83	0.33	1
401	7	16.83	0.00	22.50	0.33	1
402	5	25.83	0.00	7.67	0.00	1
403	3	4.67	0.00	7.83	0.17	1
404	6	14.33	0.00	6.17	0.00	1
405	1	46.33	0.00	16.83	0.33	1
406	4	11.50	0.00	23.33	0.00	1
407	2	18.83	0.17	7.67	0.17	1
501	3	5.50	0.67	9.50	0.00	1
502	2	9.83	5.83	12.33	0.00	1
503	6	6.67	0.33	23.17	0.67	1
504	1	24.00	0.00	12.33	0.17	1
505	4	16.33	0.17	18.83	1.00	1
506	5	10.50	0.50	12.00	0.17	1
507	7	10.83	0.67	8.17	0.33	1
601	1	25.67	14.50	16.33	0.00	1
602	3	7.83	0.00	8.67	2.00	1
603	5	24.50	0.83	3.83	0.00	1
604	2	16.83	0.17	4.67	0.00	1
605	7	30.50	0.00	7.17	0.00	1
606	6	13.83	0.00	5.00	0.00	1
607	4	33.83	0.33	7.67	0.00	1
701	3	16.17	0.83	2.50	0.00	1
702	1	51.00	1.00	9.17	0.00	1
703	6	62.83	0.50	6.17	0.17	1
704	2	18.17	1.50	12.83	1.67	1
705	5	58.83	0.00	6.33	0.00	1
706	4	54.00	0.50	13.17	0.50	1
707	7	66.67	0.00	6.83	0.00	1

<sup>1</sup> Phytotoxicity on a scale from 1 – 10 where 1 is no damage, comparable with control and 10 is complete crop kill

17.9.20						
Plot	Treatment	Parasitised <i>A. gossypii</i>	Parasitised <i>M. persicae</i>	Aphid predators	EPF <i>A.</i> <i>gossypii</i>	EPF <i>M.</i> <i>persicae</i>
101	5	0.00	0.00	0.00	0.33	2.17
102	1	0.00	0.00	0.00	0.50	0.67
103	4	0.00	0.00	0.00	0.50	5.17
104	7	0.00	0.00	0.17	0.50	5.83
105	6	0.00	0.00	0.00	0.17	0.67
106	2	0.00	0.00	0.00	0.00	0.00
107	3	0.00	0.00	0.17	0.67	1.83
201	2	0.00	0.00	0.00	0.33	0.83
202	5	0.00	0.00	0.00	0.17	1.83
203	6	0.00	0.00	0.00	0.00	0.00
204	4	0.00	0.00	0.00	0.33	0.67
205	3	0.00	0.00	0.00	0.00	0.33
206	1	0.00	0.00	0.00	0.00	1.83
207	7	0.00	0.00	0.00	0.00	6.17
301	3	0.00	0.00	0.00	0.50	0.50
302	1	0.00	0.00	0.00	2.00	6.50
303	4	0.00	0.17	0.00	1.17	3.17
304	7	0.00	0.00	0.00	0.00	0.33
305	2	0.00	0.17	0.00	1.00	3.67
306	5	0.00	0.00	0.00	0.00	0.17
307	6	0.00	0.00	0.00	0.83	4.50
401	7	0.00	0.00	0.00	0.00	0.00
402	5	0.00	0.00	0.00	0.00	0.17
403	3	0.00	0.00	0.00	0.17	1.33
404	6	0.00	0.00	0.00	0.17	0.17
405	1	0.00	0.00	0.00	0.67	1.33
406	4	0.00	0.00	0.00	1.00	1.67
407	2	0.00	0.00	0.00	0.00	0.00
501	3	0.00	0.00	0.00	0.00	2.83
502	2	0.00	0.00	0.00	0.00	0.50
503	6	0.00	0.00	0.00	0.00	2.33
504	1	0.00	0.00	0.00	0.00	0.00
505	4	0.00	0.00	0.00	0.00	0.33
506	5	0.00	0.00	0.00	0.00	0.83
507	7	0.00	0.00	0.00	0.00	0.00
601	1	0.00	0.00	0.00	3.50	0.00
602	3	0.00	0.00	0.00	0.17	0.00
603	5	0.00	0.00	0.00	0.00	0.00
604	2	0.33	0.00	0.00	0.00	0.00
605	7	0.00	0.00	0.00	0.00	0.00
606	6	0.17	0.00	0.00	0.00	0.00
607	4	0.00	0.00	0.00	0.00	0.00
701	3	0.00	0.00	0.00	0.00	0.00
702	1	0.00	0.00	0.67	0.00	0.00
703	6	0.00	0.00	0.00	0.00	0.00
704	2	0.00	0.00	0.00	0.00	0.00
705	5	0.00	0.00	0.00	0.00	0.00
706	4	0.00	0.00	0.17	0.00	0.00
707	7	0.17	0.00	0.00	0.00	0.00

24.9.20						
Plot	Treatment	Wingless <i>A.</i> <i>gossypii</i>	Winged <i>A.</i> <i>gossypii</i>	Wingless <i>M.</i> <i>persicae</i>	Winged <i>M.</i> <i>persicae</i>	Phytotoxicity <sup>1</sup>
101	5	1.67	0.00	32.17	0.33	1
102	1	5.33	0.00	34.83	0.67	1
103	4	3.50	0.33	14.50	0.17	1
104	7	5.17	0.17	53.17	1.33	1
105	6	10.33	0.00	51.50	0.17	1
106	2	11.50	0.00	21.50	0.50	1
107	3	1.00	0.00	24.17	1.17	1
201	2	2.17	0.00	43.83	1.83	1
202	5	2.00	0.00	41.00	1.67	1
203	6	4.33	0.00	13.83	0.17	1
204	4	4.83	0.00	7.00	0.17	1
205	3	3.67	0.00	9.83	0.00	1
206	1	12.17	0.00	54.83	1.00	1
207	7	6.00	0.00	40.00	0.17	1
301	3	5.00	0.00	9.67	0.00	1
302	1	2.50	0.00	108.67	0.83	1
303	4	4.17	0.00	10.67	0.33	1
304	7	28.50	0.00	45.83	0.83	1
305	2	11.17	0.17	30.17	0.50	1
306	5	9.50	0.00	24.67	0.50	1
307	6	18.00	0.00	29.83	0.50	1
401	7	39.50	0.50	29.33	0.67	1
402	5	37.50	0.17	9.83	0.00	1
403	3	8.17	0.00	4.67	0.33	1
404	6	23.00	0.67	4.67	0.50	1
405	1	73.00	0.50	23.67	1.17	1
406	4	8.17	0.17	21.00	0.67	1
407	2	20.67	0.00	10.50	0.67	1
501	3	3.33	0.00	21.00	0.00	1
502	2	24.83	0.17	34.50	2.00	1
503	6	24.33	0.00	51.00	4.67	1
504	1	36.33	0.17	29.17	0.50	1
505	4	8.83	0.00	15.17	3.17	1
506	5	24.50	1.17	37.00	2.17	1
507	7	15.17	0.83	28.83	1.83	1
601	1	31.00	0.40	47.67	0.83	1
602	3	5.00	0.00	21.33	0.83	1
603	5	48.33	0.00	30.83	2.17	1
604	2	40.00	0.00	20.50	0.33	1
605	7	47.17	0.00	36.33	0.17	1
606	6	21.17	0.33	19.83	3.67	1
607	4	33.83	1.67	20.17	2.83	1
701	3	18.50	0.83	9.00	0.00	1
702	1	93.33	0.33	36.50	1.00	1
703	6	120.67	0.67	12.33	0.50	1
704	2	14.00	0.00	23.83	3.50	1
705	5	67.17	0.00	6.00	0.17	1
706	4	31.67	0.17	38.17	0.67	1
707	7	117.17	0.33	34.33	1.50	1

<sup>1</sup> Phytotoxicity on a scale from 1 – 10 where 1 is no damage, comparable with control and 10 is complete crop kill

24.09.20						
Plot	Treatment	Parasitised <i>A. gossypii</i>	Parasitised <i>M. persicae</i>	Aphid predators	EPF <i>A.</i> <i>gossypii</i>	EPF <i>M.</i> <i>persicae</i>
101	5	0.00	0.00	0.00	0.50	2.00
102	1	0.00	0.00	0.00	0.83	5.33
103	4	0.00	0.00	0.00	1.33	11.17
104	7	0.00	0.00	0.00	1.00	11.17
105	6	0.00	0.00	0.00	2.17	12.17
106	2	0.00	0.00	0.00	0.00	0.50
107	3	0.00	0.00	0.00	0.17	3.00
201	2	0.00	0.00	0.17	0.33	2.17
202	5	0.00	0.00	0.00	0.33	2.17
203	6	0.00	1.67	0.00	0.00	0.00
204	4	0.00	0.00	0.00	0.00	0.33
205	3	0.00	0.00	0.00	0.17	0.00
206	1	0.00	0.00	0.00	0.83	1.83
207	7	0.00	0.00	0.17	3.83	10.50
301	3	0.00	0.00	0.00	0.50	0.17
302	1	0.00	0.00	0.00	2.83	16.67
303	4	0.00	0.00	0.00	0.33	3.00
304	7	0.00	0.00	0.00	0.50	0.17
305	2	0.00	0.00	0.00	1.17	4.50
306	5	0.00	0.00	0.17	0.00	0.83
307	6	0.00	0.00	0.00	1.33	4.50
401	7	0.00	0.00	0.00	0.50	2.50
402	5	0.00	0.00	0.00	0.17	0.00
403	3	0.00	0.00	0.00	0.00	0.50
404	6	0.00	0.00	0.00	0.00	0.17
405	1	0.00	0.00	0.00	0.50	5.00
406	4	0.00	0.00	0.00	0.83	3.83
407	2	0.00	0.00	0.00	0.00	0.00
501	3	0.00	0.00	0.00	0.00	4.17
502	2	0.00	0.00	0.00	0.17	0.17
503	6	0.00	0.67	0.17	1.00	6.50
504	1	0.00	0.00	0.00	0.17	0.50
505	4	0.00	0.00	0.00	0.33	2.00
506	5	0.00	0.00	0.00	0.00	4.83
507	7	0.00	0.00	0.00	0.17	0.67
601	1	0.00	0.33	0.00	5.33	4.50
602	3	0.00	0.33	0.00	0.50	0.00
603	5	0.00	0.00	0.00	0.00	0.00
604	2	0.00	0.00	0.00	1.67	0.50
605	7	0.00	0.00	0.00	0.50	0.67
606	6	0.00	0.00	0.00	0.00	0.00
607	4	0.00	0.00	0.17	0.00	0.00
701	3	0.00	0.00	0.00	1.50	0.17
702	1	0.00	0.00	0.00	2.50	7.67
703	6	0.00	0.00	0.00	2.33	0.00
704	2	0.00	0.00	0.00	0.33	0.00
705	5	0.00	0.33	0.00	0.33	0.00
706	4	0.00	0.00	0.00	2.83	0.50
707	7	0.00	0.00	0.00	0.50	0.00

1.10.20						
Plot	Treatment	Wingless <i>A.</i> <i>gossypii</i>	Winged <i>A.</i> <i>gossypii</i>	Wingless <i>M.</i> <i>persicae</i>	Winged <i>M.</i> <i>persicae</i>	Phytotoxicity <sup>1</sup>
101	5	2.83	0.00	50.83	0.50	1
102	1	6.83	0.17	59.33	0.33	1
103	4	2.67	0.00	20.17	0.00	1
104	7	10.17	0.00	51.17	0.50	1
105	6	9.50	0.00	52.50	0.17	1
106	2	15.17	0.00	23.50	0.00	1
107	3	1.50	0.00	24.17	0.33	1
201	2	1.33	0.17	66.17	0.83	1
202	5	2.33	0.00	57.83	0.17	1
203	6	6.33	0.00	28.83	0.33	1
204	4	3.83	0.00	9.67	0.00	1
205	3	2.17	0.00	20.33	0.00	1
206	1	11.00	0.00	61.83	0.00	1
207	7	4.67	0.00	41.33	0.00	1
301	3	2.67	0.00	12.50	0.17	1
302	1	0.83	0.00	60.33	0.00	1
303	4	4.00	0.00	18.17	0.33	1
304	7	22.33	0.00	72.83	1.50	1
305	2	7.33	0.17	43.67	0.33	1
306	5	7.00	0.33	26.33	0.67	1
307	6	18.17	0.00	30.67	0.50	1
401	7	27.50	4.67	20.50	9.00	1
402	5	19.67	2.33	8.67	2.00	1
403	3	4.17	1.67	5.33	1.33	1
404	6	15.33	0.83	10.67	1.33	1
405	1	23.17	10.83	27.83	8.00	1
406	4	4.33	1.83	16.00	3.33	1
407	2	12.67	3.67	13.17	4.17	1
501	3	10.17	2.17	21.33	2.33	1
502	2	39.83	8.67	72.67	9.83	1
503	6	17.33	6.17	35.17	12.83	1
504	1	22.83	1.00	32.50	2.83	1
505	4	8.83	3.33	18.67	6.33	1
506	5	22.50	5.00	36.00	7.83	1
507	7	26.00	8.00	36.33	8.33	1
601	1	30.33	7.83	23.33	4.00	1
602	3	17.17	5.17	31.00	3.17	1
603	5	54.17	1.17	42.50	7.00	1
604	2	27.67	0.83	26.83	2.33	1
605	7	38.83	5.00	44.17	6.00	1
606	6	19.67	2.50	34.50	3.17	1
607	4	16.50	1.17	29.00	4.83	1
701	3	15.50	0.00	15.67	0.00	1
702	1	83.67	6.83	26.50	4.17	1
703	6	87.17	9.33	20.67	3.50	1
704	2	23.00	10.17	25.83	1.67	1
705	5	26.83	0.00	4.00	0.00	1
706	4	31.50	3.00	28.83	1.17	1
707	7	96.50	14.00	41.83	8.00	1

<sup>1</sup> Phytotoxicity on a scale from 1 – 10 where 1 is no damage, comparable with control and 10 is complete crop kill

1.10.20						
Plot	Treatment	Parasitised <i>A. gossypii</i>	Parasitised <i>M.</i> <i>persicae</i>	Aphid predators	EPF <i>A.</i> <i>gossypii</i>	EPF <i>M.</i> <i>persicae</i>
101	5	0.00	0.00	0.17	0.83	7.50
102	1	0.00	0.00	0.33	2.67	9.50
103	4	0.17	0.17	0.33	1.50	7.83
104	7	0.00	0.50	0.00	5.00	18.33
105	6	0.00	0.00	0.00	5.00	19.50
106	2	0.00	0.17	0.00	0.50	4.33
107	3	0.00	0.00	0.00	0.00	5.33
201	2	0.00	0.17	0.00	1.50	7.83
202	5	0.00	0.00	0.00	1.00	14.17
203	6	1.83	0.00	0.00	0.00	0.00
204	4	0.00	0.00	0.00	0.00	0.17
205	3	0.00	0.00	0.00	0.00	0.00
206	1	0.00	0.00	0.00	3.33	10.17
207	7	0.00	0.00	0.00	3.33	12.17
301	3	0.00	0.00	0.00	0.00	0.17
302	1	0.00	0.33	0.00	6.83	29.67
303	4	0.00	0.00	0.17	0.00	5.50
304	7	0.00	0.00	0.00	0.17	5.17
305	2	0.00	0.00	0.00	1.83	13.33
306	5	0.00	0.00	0.00	0.83	3.33
307	6	0.00	0.00	0.00	3.17	11.33
401	7	0.00	0.00	0.00	4.83	5.17
402	5	0.00	0.00	0.00	1.33	2.17
403	3	0.00	0.00	0.00	0.00	0.00
404	6	0.00	0.00	0.00	0.00	0.00
405	1	0.00	0.00	0.00	9.33	10.00
406	4	0.00	0.00	0.00	2.33	5.67
407	2	0.00	0.00	0.00	0.00	0.33
501	3	0.00	0.00	0.00	8.17	5.67
502	2	0.00	0.00	0.00	7.17	5.50
503	6	0.00	0.00	0.00	5.67	8.83
504	1	0.00	0.00	0.00	2.00	3.67
505	4	0.00	0.00	0.00	0.50	2.00
506	5	0.00	0.00	0.00	3.67	6.00
507	7	0.00	0.00	0.00	5.17	9.50
601	1	0.00	0.00	0.00	15.50	15.17
602	3	0.00	0.00	0.00	0.17	0.50
603	5	0.00	0.00	0.00	0.00	0.00
604	2	0.00	0.00	0.00	5.83	5.67
605	7	0.00	0.00	0.00	6.33	8.83
606	6	0.00	0.00	0.00	3.17	3.67
607	4	0.00	0.00	0.00	0.00	0.00
701	3	0.00	0.00	0.00	0.00	0.00
702	1	0.00	0.00	0.00	17.67	4.33
703	6	0.00	0.00	0.00	0.00	0.00
704	2	0.00	0.00	0.00	2.17	0.00
705	5	0.00	0.00	0.00	0.33	0.00
706	4	0.00	0.00	0.00	0.00	0.00
707	7	0.00	0.00	0.00	5.33	1.67

f. Trial design

Door

Block 7	Plot	701	702	703	704	705	706	707
	Trt	3	1	6	2	5	4	7
Block 6	Plot	601	602	603	604	605	606	607
	Trt	1	3	5	2	7	6	4
Block 5	Plot	501	502	503	504	505	506	507
	Trt	3	2	6	1	4	5	7
Block 4	Plot	401	402	403	404	405	406	407
	Trt	7	5	3	6	1	4	2
Block 3	Plot	301	302	303	304	305	306	307
	Trt	3	1	4	7	2	5	6
Block 2	Plot	201	202	203	204	205	206	207
	Trt	2	5	6	4	3	1	7
Block 1	Block 1	101	102	103	104	105	106	107
	Trt	5	1	4	7	6	2	3

Code	Treatment
T1	Untreated (-ve control)
T2	Mainman (+ve control industry standard)
T3	AHDB9968
T4	AHDB9921
T5	AHDB9920
T6	AHDB9919
T7	AHDB9918

g. ORETO certificate.



*Certificate of*

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

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*This certifies that*

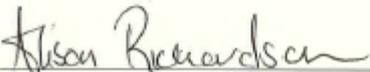
**RSK ADAS Ltd**

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  
Stored Crops  
Biologicals and Semiochemicals**

**Date of issue: 1 June 2018**  
**Effective date: 18 March 2018**  
**Expiry date: 17 March 2023**

**Signature**   
Authorised signatory

<b>Certification Number</b> ORETO 409
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Chemicals Regulation Division

 Department of  
Agriculture and  
Rural Development