

# SCEPTREPLUS

## Final Trial Report

<b>Trial code:</b>	SP37
<b>Title:</b>	Control of Downy Mildew (caused by <i>Hyaloperonospora brassicae</i> ) in propagated brassica crops
<b>Crop:</b>	Cauliflower Also applicable to other propagated brassica crops
<b>Target:</b>	Downy Mildew (caused by <i>Hyaloperonospora brassicae</i> )
<b>Lead researcher:</b>	Kirsty Wright
<b>Organisation:</b>	Stockbridge Technology Centre
<b>Period:</b>	May-July 2018
<b>Report date:</b>	09 November 2018
<b>Report author:</b>	Kirsty Wright
<b>ORETO Number: (certificate should be attached)</b>	ORETO 372

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.



...07/11/18.....

Date

.....  
Authors signature

## Trial Summary

### Introduction

*Hyaloperonospora brassicae* causes the rapid loss of infected brassica cotyledons and subsequent lesions on true leaves can cause stunting and reduced quality of transplants. Systemic infection in cauliflowers causes discolouration in the curds and in cabbage and Brussels sprouts lesions may penetrate into several leaf layers causing reduced crop quality. The pathogen has a wide brassica host range and spores are quick to infect, as long as leaf surfaces are wet; such as in irrigated glasshouse propagation crops. The year-round production of transplants enables the continual spread of spores to young crops. Growers have some approved products available to them, but most of these fall into the same FRAC group which is a cause for concern. This trial sought to identify the best products already approved for use, and to test some alternative products, for their efficacy against brassica downy mildew.

### Methods

Cauliflower cv. Graffiti was sown into one 308 module tray per plot, replicated 4 times per treatment. Treatments were applied at cotyledon stage and the crop inoculated with a spore suspension of *Hyaloperonospora brassicae* two days later. The crop was kept humid to ensure ideal conditions for disease spread. Treatments were reapplied as per the product label, EAMU or manufacturers guidelines. Assessments commenced once downy mildew symptoms (leaf spots, yellowing cotyledons) were observed and continued until the crop was approximately six weeks old and ready for transplanting. A count of the number of plants with disease symptoms in the central 30 cells of each module tray was done at each assessment and subsequently converted to a percentage.

### Results

Trt No.	Treatment code (and name where approved)	% of plants with downy mildew leaf spots (back-transformed data)		
		11.07.18	17.07.18	24.07.18
1	Untreated	14.28	12.26	14.40
2	AHDB 9942 (Peraat)	5.47	15.08	11.13
3	AHDB 9882 (Infinito)	0.68	1.98	1.68
4	AHDB 9881 (Proplant)	0.48	5.88	1.92
5	AHDB 9941 (Previcur Energy)	4.32	8.92	3.63
6	AHDB 9880	10.60	22.07	23.26
7	AHDB 9939	0.47	1.49	0.69
8	AHDB 9883	0.46	1.08	2.92
9	AHDB 9962	0.00	0.68	1.42
10	AHDB 9879	1.10	3.22	1.43
	Not significantly different from untreated control (p>0.05)			
	Significantly different from untreated control (p<0.05)			

### Conclusions

All products tested were crop-safe during the life of the trial. The standard product (Peraat) did not perform well in this trial, potentially due to pathogen resistance, but three other approved products (Infinito, Proplant and Previcur Energy) all significantly reduced downy mildew symptoms. One product (AHDB 9880) appeared to increase plant susceptibility to disease, giving higher levels of leaf spotting than the untreated. Two products (AHDB 9939 and AHDB 9883) are already approved on other crops in

the UK and gave good control of disease symptoms, whilst two novel products (AHDB 9962 and AHDB 9879) also significantly reduced disease levels.

**Take home message**

- Infinito and Proplant were the most effective approved products in the trial. Infinito combines two modes of action and is therefore a good choice of product.
- Paraat should be used with caution.
- Some of the close to market alternatives tested show good potential for disease control and possible approval in the near future.

## Objectives

To assess a selection of approved and novel fungicides for activity against downy mildew (caused by *Hyaloperonospora brassicae*) of brassicas grown in propagation.

## 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
PP 1/152(3)	Design and analysis of efficacy evaluation trials	None
PP 1/135(3)	Phytotoxicity assessment	None
PP 1/181(3)	Conduct and reporting of efficacy evaluation trials including GEP	None

There were no deviations from EPPO guidance.

## Test site

Item	Details
Location address	Glasshouse F12, Stockbridge Technology Centre, Stockbridge House, Cawood, Selby YO8 3TZ
Crop	Cauliflower ( <i>Brassica oleracea var. botrytis</i> , BRSOB)
Cultivar	Graffiti
Soil or substrate type	Levington F2+S
Agronomic practice	Benched crop, watered overhead, liquid feed when required, no insecticides or fungicides other than test treatments applied.
Prior history of site	Propagation unit, wide variety of crops.

## Trial design

Item	Details
Trial design:	Randomised complete block
Number of replicates:	4
Row spacing:	n/a
Plot size: (w x l)	0.4 x 0.6m
Plot size: (m <sup>2</sup> )	0.24m <sup>2</sup>
Number of plants per plot:	308 (of which 30 were assessed)
Leaf Wall Area calculations	N/A

## Treatment details

Trt. No.	AHDB Code	Active substance	Product name/ manufacturers code	Formulation batch number	Content of active substance in product	Formulation type
1	-	Untreated (water only)				
2	AHDB 9942	Dimethomorph	Paraat	2351-01	50%	WP
3	AHDB 9882	Fluopicolide + propamocarb hydrochloride	Infito	N/A	62.5 g/l + 625 g/l	SC
4	AHDB 9881	Propamocarb hydrochloride	Proplant	17260400	722 g/l (62.9% w/w)	SL
5	AHDB 9941	Fosetyl-aluminium + propamocarb hydrochloride	Previcur Energy	EM4L01904 1	530 g/l + 310 g/l	SL
6	AHDB 9880	N/D	N/D	N/D	N/D	SP
7	AHDB 9939	N/D	N/D	N/D	N/D	SC
8	AHDB 9883	N/D	N/D	N/D	N/D	SC
9	AHDB 9962	N/D	N/D	N/D	N/D	OD
10	AHDB 9879	N/D	N/D	N/D	N/D	SC

## 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	-			CDGHIJ
2	AHDB 9942	180 g/ha	0.36 kg/ha	CD
3	AHDB 9882	100 g/ha + 1000 g/ha	1.6 l/ha	C
4	AHDB 9881	36100 g/ha	50.0 l/ha	A
5	AHDB 9941	15900 g/ha + 9300 g/ha	30.0 l/ha (3ml/m <sup>2</sup> )	AF
6	AHDB 9880	2550 g/ha	3.0 kg/ha	CDGI
7	AHDB 9939	150 g/ha	0.6 l/ha	C
8	AHDB 9883	100 g/ha	0.625l/ha	CDH
9	AHDB 9962	15 g/ha + 35 g/ha	0.5 l/ha	CDHJ
10	AHDB 9879	157.5 g/ha + 105 g/ha	0.7 l/ha	BE

## Application details

	Application A	Application B	Application C	Application D	Application E
Application date	20.06.18	20.06.18	20.06.18	28.06.18	04.07.18
Time of day	13:30-14:30	13:30-14:30	13:30-14:30	13:00-13:30	11:00-12:00
Crop growth stage (average BBCH)	10	10	10	11	11
Crop height (cm)	N/A	N/A	N/A	N/A	N/A
Crop coverage (%)	100	>90	>90	>90	>90
Application Method	Spray	Spray	Spray	Spray	Spray
Application Placement	Drench	Foliar	Foliar	Foliar	Foliar
Application equipment	OPS	OPS	OPS	OPS	OPS
Nozzle pressure	2 bar				
Nozzle type	Flat Fan				
Nozzle size	10 F110	01 F80	01 F80	01 F80	01 F80
Application water volume/ha	20000	200	600	600	200
Temp of air - shade (°C)	24.8	24.8	24.8	32.7	23.5
Relative humidity (%)	N/A	N/A	N/A	32.3	58.6
Wind speed range (m/s)	0	0	0	0	0
Dew presence (Y/N)	N	N	N	N	N
Temperature of soil - 2-5 cm (°C)	N/A	N/A	N/A	N/A	N/A
Wetness of soil - 2-5 cm	N/A	N/A	N/A	N/A	N/A
Cloud cover (%)	60	60	60	0	100

	Application F	Application G	Application H	Application I	Application J
Application date	04.07.18	04.07.18	06.07.18	11.07.18	16.07.18
Time of day	11:00-12:00	11:00-12:00	10:00-10:30	13:30-14:00	10:00-10:30
Crop growth stage (average BBCH)	11	11-12	11-12	11-12	12-13
Crop height (cm)	N/A	N/A	N/A	N/A	N/A
Crop coverage (%)	100	>90	>90	>90	>90
Application Method	Spray	Spray	Spray	Spray	Spray
Application Placement	Foliar	Foliar	Foliar	Foliar	Foliar
Application equipment	OPS	OPS	OPS	OPS	OPS
Nozzle pressure	2 bar				
Nozzle type	Flat Fan				
Nozzle size	10 F110	01 F80	01 F80	01 F80	01 F80
Application water volume/ha	20000	600	600	600	600
Temp of air - shade (°C)	23.5	23.5	26.5	32.6	25.8
Relative humidity (%)	58.6	58.6	48.7	34.9	55.1
Wind speed range (m/s)	0	0	0	0	0
Dew presence (Y/N)	N	N	N	N	N
Temperature of soil - 2-5 cm (°C)	N/A	N/A	N/A	N/A	N/A
Wetness of soil - 2-5 cm	N/A	N/A	N/A	N/A	N/A
Cloud cover (%)	100	100	50	60	90

## 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
Downy Mildew	<i>Hyaloperonospora brassicae</i>	HYPERBR	Nil	Moderate (28% of untreated plants with leaf spots)	Moderate (17% of untreated plants with leaf spots)

## Assessment details

	Evaluation Timing (DA)*			
Evaluation date	After conventional insecticides	Crop Growth Stage (BBCH)	Evaluation type (efficacy, phytotox)	Assessment
03.07.18	Varies according to treatment	11	Establishment	Count of plants in assessment area of each plot
03.07.18		11	Efficacy	Count of plants with yellowed cotyledons
03.07.18		11	Efficacy	Count of plants with senesced cotyledons
03.07.18		11	Efficacy	Count of plants with downy mildew leaf spots (cotyledons)
11.07.18		11-12	Efficacy	Count of plants with retained cotyledons
11.07.18		11-12	Efficacy	Count of plants with downy mildew leaf spots (true leaves)
17.07.18		12-13	Efficacy	Count of plants with downy mildew leaf spots (true leaves)
24.07.18		13	Efficacy	Count of plants with downy mildew leaf spots (true leaves)

\* DA – days after previous application

The central 30 cells in each tray were assessed at each timing and the count of infected seedlings converted to a percentage using the plant count carried out at the first assessment.

## Statistical analysis

This trial was designed as a randomized complete block with 10 treatments and 4 replicates. Where suitable for analysis, data were analysed by ANOVA using the Minitab (v17) program by David George at STC. Prior to analysis data were checked for normality and homoscedasticity, being transformed, if required, using a log (n+1) transformation. Where ANOVA reported a statistically significant effect of treatment, *post-hoc* testing was conducted by calculating one-tailed LSDs to allow treatments to be compared against the untreated control for demonstration of efficacy.

## Results

### Phytotoxicity

No symptoms of phytotoxicity were seen with any of the treatments used in this trial.

### Efficacy

Analysis of a selection of assessments is shown in Table 1 below and percent reduction in leaf spots is shown in Table 2. A selection of assessments are also presented in the graphs below.

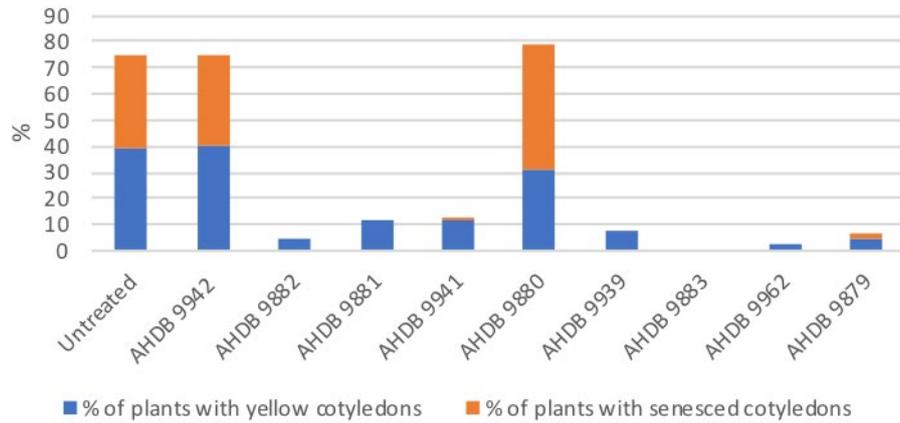
**Table 1: Analysis of efficacy assessments**

		3.7.18	11.7.18	17.7.18	24.7.18
Trt No.		% plants with yellowed cotyledons	% of plants with downy mildew leaf spots on true leaves	% of plants with downy mildew leaf spots on true leaves	% of plants with downy mildew leaf spots on true leaves
1	Untreated	39.6791	2.7266	2.5846	2.7345
2	AHDB 9942	40.0702	1.8666	2.7775	2.4959
3	AHDB 9882	4.4643	0.5166	1.0925	0.9871
4	AHDB 9881	12.0017	0.3945	1.9293	1.0729
5	AHDB 9941	11.4914	1.6721	2.2949	1.5319
6	AHDB 9880	31.4667	2.4507	3.1386	3.1888
7	AHDB 9939	7.3082	0.3871	0.9122	0.5243
8	AHDB 9883	0.0000	0.3800	0.7342	1.3671
9	AHDB 9962	2.5000	0.0000	0.5166	0.8823
10	AHDB 9879	4.3144	0.7397	1.4406	0.8892
	F / H	8.3200	4.08	3.33	3.35
	P	0.0000	0.002	0.006	0.006
	df	30.0000	30	30	30
	lsd	10.6068	0.9474	0.9992	0.9769
	tcrit(df) one tailed	1.6970	1.697	1.697	1.697
	MSE	117.2000	0.9351	1.04	0.9942
	LSD	10.6068	0.9474	0.9992	0.9769
			data transformed : ln (x+1) -still not normal p = 0.027	data transformed: ln (x+1)	data transformed: ln (x+1)

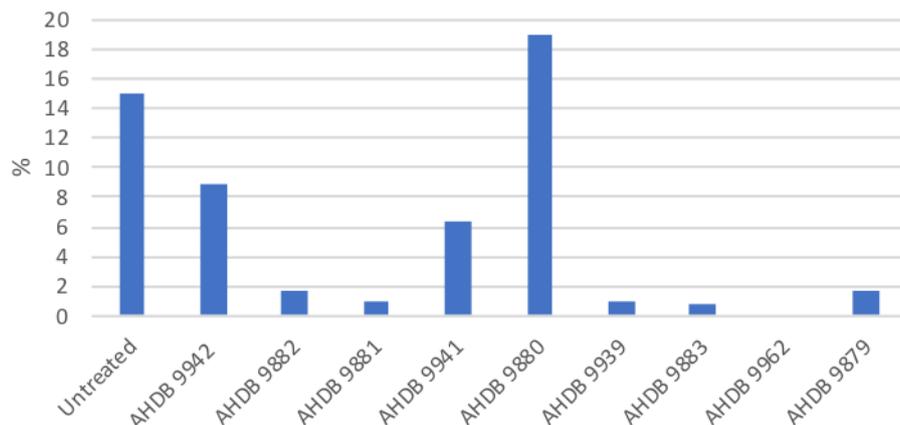
**Table 2: Percent reduction in downy mildew leaf spots**

Trt No.	Treatment code (and name where approved)	% reduction in downy mildew leaf spots (based on raw data)		
		11.07.18	17.07.18	24.07.18
1	Untreated			
2	AHDB 9942	40.3	-9.5	23.0
3	AHDB 9882	88.5	64.7	80.1
4	AHDB 9881	93.6	37.5	78.4
5	AHDB 9941	57.6	30.4	63.1
6	AHDB 9880	-26.5	-63.4	-52.2
7	AHDB 9939	93.8	81.2	89.8
8	AHDB 9883	94.0	69.4	74.9
9	AHDB 9962	100.0	88.2	85.5
10	AHDB 9879	88.7	63.9	85.3
	Not significantly different from untreated control (p>0.05)			
	Significantly different from untreated control (p<0.05)			

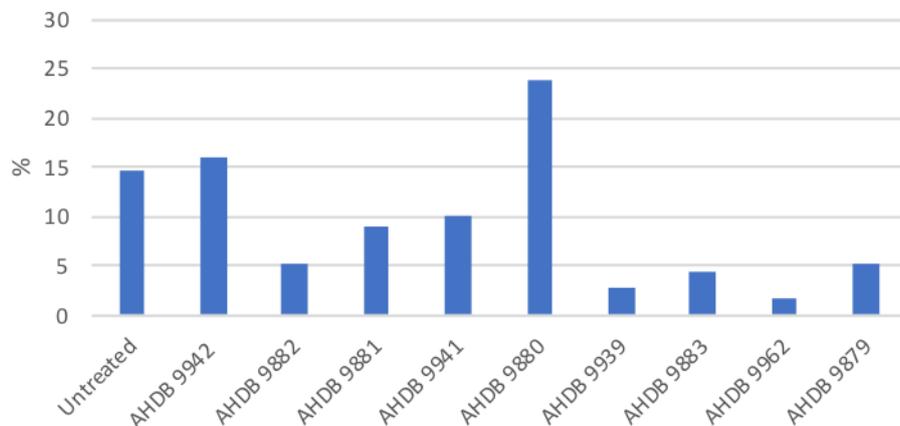
% of plants with yellowed or senesced cotyledons  
03.07.18

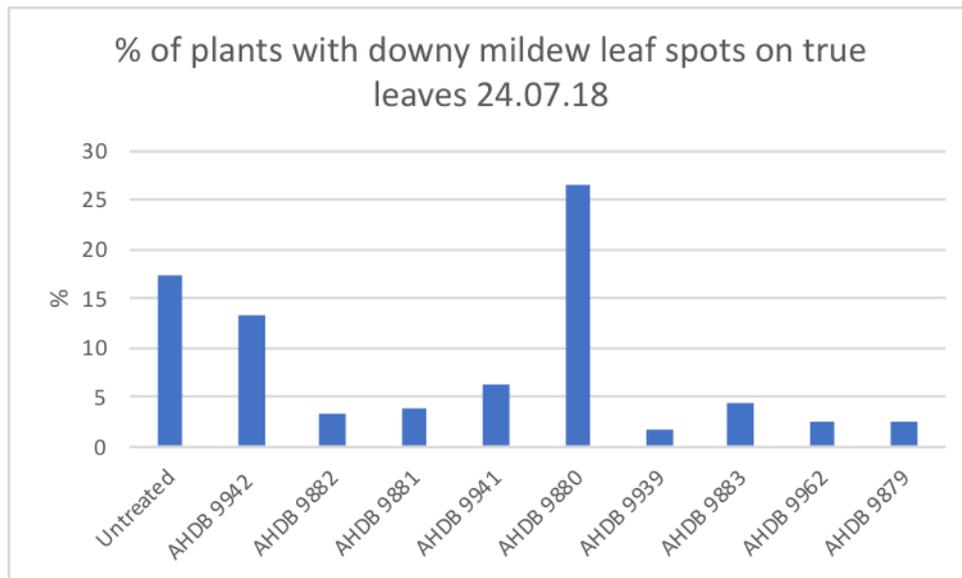


% of plants with downy mildew leaf spots on true leaves 11.07.18



% of plants with downy mildew leaf spots on true leaves 17.07.18





## Discussion

The trial was conducted as described in the protocol with the following exceptions:

- Treatment 5 (AHDB 9941) was first applied at cotyledon stage and not pre-emergence.
- Results were analysed using Minitab instead of ARM software.

Disease levels were moderate throughout the trial, with an average of 17% of untreated plants showing downy mildew leaf spots at the end of the trial.

No phytotoxicity was seen with any of the products used, at the rates tested.

Only two of the nine products tested did not give any significant control of disease symptoms. AHDB 9942 (Paraat) was included as a standard product following advice from growers. It did not achieve satisfactory control of disease in the trial although other products did. Feedback from growers suggests that they have observed a good eradicant effect when using Paraat and we may have seen a better effect using the product after inoculation. However, this is contrary to label advice for control of other pathogens and the cause of poor control may have been reduced sensitivity of the *H. brassicae* isolate used.

AHDB 9880 actually seemed to increase disease levels relative to the untreated control. Upon application of this product, leaves appeared wetter following application compared with other products and this may have promoted pathogen infection.

Of the remaining products tested, three are already approved for use on brassica seedlings; two of these, namely AHDB 9882 (Infinito) and AHDB 9881 (Proplant) gave good control of downy mildew and the other (AHDB 9941, Previcur Energy) gave moderate control.

AHDB 9939 gave the best control of downy mildew symptoms in the trial and AHDB 9883 also gave good control. Both products are already approved on other crops in the UK, making them good candidates for approvals if appropriate residue data can be generated. Two novel fungicides (AHDB 9962 and AHDB 9879) were also used in the trial and also gave a good reduction in disease symptoms.

Fungicide resistance management is an important factor in the control of downy mildew infections. Growers should aim to utilise fungicides with different modes of action in order to minimise the risk of resistance in the pathogen population as recommended by the Fungicide Resistance Action Committee (FRAC). The table below shows the FRAC groups codes for the products tested. Of the four currently approved products (Treatments 2-5), only 3 treatments were effective in this trial and all contain propamocarb hydrochloride (FRAC code 28). This highlights the requirement for additional products to be made available to brassica propagators.

Treatment No.	AHDB Code	FRAC Groups
2	AHDB 9942	40
3	AHDB 9882	43 + 28
4	AHDB 9881	28
5	AHDB 9941	33 + 28
6	AHDB 9880	-
7	AHDB 9939	40
8	AHDB 9883	21
9	AHDB 9962	49 + 40
10	AHDB 9879	22 + 4

## Conclusions

- Disease levels were moderate in the untreated plots.
- No products caused phytotoxicity.
- The standard product did not work well, with very little control of downy mildew leaf spots seen. This may be because of reduced sensitivity of the pathogen strain to the product or, potentially, because of suboptimal application timing.
- All but one test product gave moderate or good control of disease symptoms.

## Acknowledgements

We would like to thank AHDB and the participating crop protection companies for project funding, Plant Propagators Ltd for all their input and advice and Allan Cropley for supplying infected leaf material.

## Appendix A: Crop Diary

Date	Action
11.06.18	44x 308 trays sown with Cauliflower cv. Graffiti in F2+S
	Watering and feed as required. No other pesticides applied

## Appendix B: Trial Diary

Date	Action
20.06.18	Applications A, B and C made
22.06.18	Crop moved to potting shed and inoculum applied. Crop covered with polythene and left overnight
23.06.18	Crop returned to benches in glasshouse F12
28.06.18	Application D made
03.07.18	Assessments of efficacy
04.07.18	Applications E, F and G made
06.07.18	Application H made
11.07.18	Assessments of efficacy
11.07.18	Application I made
16.07.18	Application J made
17.07.18	Assessments of efficacy
24.07.18	Assessments of efficacy

## Appendix C: Photos



Trial setup in glasshouse. Note capillary matting and shade netting to encourage high humidity



Downy mildew leaf spots on true leaves

The series of photos below shows the difference in cotyledon effects caused by downy mildew.

Treatment 1:



Treatment 2:



Treatment 3:



Treatment 4:



Treatment 5:



Treatment 6:



Treatment 7:



Treatment 8:



Treatment 9:



Treatment 10:



## Appendix D: Climatological Data

	average temperature (°C)	average RH (%)
11/06/2018	18.63	n/a
12/06/2018	19.92	n/a
13/06/2018	19.72	n/a
14/06/2018	18.03	n/a
15/06/2018	17.53	n/a
16/06/2018	17.8	n/a
17/06/2018	19.39	n/a
18/06/2018	19	n/a
19/06/2018	19.62	n/a
20/06/2018	17.8	n/a
21/06/2018	17.53	n/a
22/06/2018	19.9	n/a
23/06/2018	18.18	n/a
24/06/2018	20.73	n/a
25/06/2018	21.05	n/a
26/06/2018	20.32	n/a
27/06/2018	20.1	n/a
28/06/2018	20.5	65.71
29/06/2018	18.57	66.22
30/06/2018	20.2	72
01/07/2018	20.4	64.7
02/07/2018	20.4	67.75
03/07/2018	19.82	72.82
04/07/2018	18.93	73
05/07/2018	20.43	75
06/07/2018	21.72	69
07/07/2018	20.89	59
08/07/2018	22.13	67
09/07/2018	19.8	78
10/07/2018	18.32	69
11/07/2018	20.53	69.7
12/07/2018	20.2	77
13/07/2018	21.12	64.18
14/07/2018	22	57
15/07/2018	21.62	62
16/07/2018	20.68	76.2
17/07/2018	17.99	65
18/07/2018	20.4	70
19/07/2018	18.82	58
20/07/2018	20.1	71.83
21/07/2018	19.3	71
22/07/2018	22.3	74
23/07/2018	22.9	74.82
24/07/2018	19.07	71

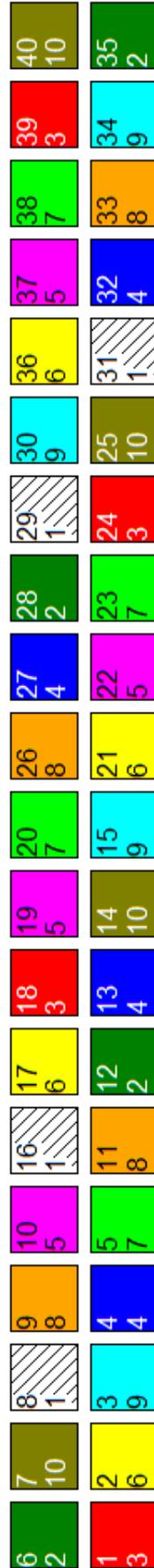
## Appendix E: Raw Data

		3.7.18	3.7.18	3.7.18	3.7.18	11.7.18	11.7.18	17.7.18	24.7.18
plot	Treatment number	total number of plants (in 30 cells)	number of plants with yellowed cotyledons	number of plants with DM spots on cotyledons	number of plants with completely senesced cotyledons	number of plants with cotyledons retained	number of plants with DM lesions on true leaves	number of plants with DM lesions on true leaves	number of plants with DM lesions on true leaves
8	1	27	6	4	3	0	6	1	3
16	1	29	13	8	14	0	3	6	2
29	1	28	14	12	11	0	3	6	4
31	1	24	10	7	10	0	4	3	9
6	2	29	8	0	5	0	0	3	3
12	2	29	11	5	11	0	2	4	2
28	2	27	20	14	6	0	5	7	8
35	2	29	6	0	18	1	3	4	2
1	3	28	5	0	0	2	0	0	1
18	3	29	0	0	0	0	2	5	3
24	3	25	0	0	0	3	0	0	0
39	3	30	0	0	0	2	0	1	0
4	4	29	1	0	0	0	0	4	0
13	4	30	2	0	0	2	0	0	0
27	4	27	4	0	0	0	0	3	2
32	4	26	6	3	0	0	1	3	2
10	5	27	1	0	0	0	3	1	1
19	5	28	3	0	0	0	2	5	5
22	5	24	5	2	0	0	0	2	1
37	5	28	3	2	1	3	2	3	0
2	6	27	5	2	4	0	0	5	6

		3.7.18	3.7.18	3.7.18	3.7.18	11.7.18	11.7.18	17.7.18	24.7.18
plot	Treatment number	total number of plants (in 30 cells)	number of plants with yellowed cotyledons	number of plants with DM spots on cotyledons	number of plants with completely senesced cotyledons	number of plants with cotyledons retained	number of plants with DM lesions on true leaves	number of plants with DM lesions on true leaves	number of plants with DM lesions on true leaves
17	6	30	10	14	18	0	8	4	3
21	6	28	13	10	14	1	7	11	13
36	6	29	8	6	19	1	7	7	8
5	7	28	0	0	0	1	0	1	2
20	7	27	0	0	0	5	1	2	0
23	7	27	5	0	0	2	0	0	0
38	7	28	3	0	0	5	0	0	0
9	8	29	0	0	0	2	0	0	3
11	8	28	0	0	0	4	1	5	1
26	8	27	0	0	0	2	0	0	0
33	8	28	0	0	0	1	0	0	1
3	9	30	0	0	0	5	0	0	2
15	9	29	0	0	0	1	0	2	1
30	9	30	3	0	0	1	0	0	0
34	9	28	0	0	0	0	0	0	0
7	10	30	2	0	1	1	1	0	2
14	10	29	1	0	0	1	1	4	0
25	10	27	0	0	0	3	0	1	0
40	10	28	2	2	2	1	0	1	1

# Appendix F: Trial Layout

Trt	Code	Description
1	CHK	Untreated
2		AHDB 9942
3		AHDB 9882
4		AHDB 9881
5		AHDB 9941
6		AHDB 9880
7		AHDB 9939
8		AHDB 9883
9		AHDB 9962
10		AHDB 9879



## Appendix G: ORETO Certificate



# Certificate of

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

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*This certifies that*

**Stockbridge Technology Centre**

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  
Biologicals and Semiochemicals  
Stored Crops**

**Date of issue:** 19 July 2016  
**Effective date:** 1 April 2016  
**Expiry date:** 31 March 2021

**Signature**

  
*Authorised Signatory*

Certification Number

ORETO 372



Chemicals Regulation Division



Department of  
**Agriculture and  
Rural Development**