CP 205 AHDB Horticulture Efficacy Trials 2022
Final Trial Report

Work package:	WP 14
Title:	SCEPTREplus - Control of fungal leaf spots of hebe
Crop	*Hebe rakaiensis*
Target	Fungal leaf spots; Septoria and Stemphylium
Lead researcher:	Erika Wedgwood
Organisation:	ADAS
Period:	18/10/2022 – 13/12/2022
Report date:	27/01/2023
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ORETO Number:	23-05

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

Date 27 January 2023

Author's signature E. F. Wedgwood

Trial Summary

Introduction

The range of products currently available for the control of fungal leaf spot is limited. Issues with fungal leaf spot have been noted at commercial nurseries in the past**.** *Hebe spp.* are hardy nursery stock plants that are susceptible to leafspot caused by both Septoria and Stemphylium.

The leafspot lesions cause loss of marketability. Stemphylium spots are dark, 1-3mm in diameter, and do not merge. Septoria spots also start dark but become larger and develop a pale centre with tiny spore bodies, and the lesions do tend to merge.

Septoria was regarded as the predominant pathogen, but a survey in 2008 found that Stemphylium was more prevalent. Both can be present on the same plant.

Stemphylium favours warm, wet weather and is most prevalent during the summer and autumn. Septoria is also spread via watersplash and so is more severe in wet weather.

The objective of these trials was to identify crop-safe and efficient products for use as control agents against fungal leaf spot, to seek to expand the range of plant protection product options available to growers.

Methods

The trial was sited in a glasshouse at a commercial nursery with a history of leafspots and set up on 18 October 2022 with *Hebe rakaiensis* liners from that nursery in 105 mm diameter pots, with 15 plants per plot. There were nine treatments, including an untreated which received a water spray. Two commercial standards were used: Signum (boscalid plus pyraclostrobin) and Luna Privilege (fluopyram). The experimental products comprised a conventional chemical fungicide (AHDB 9714), two microbial bioprotectants (AHDB 9713 & 9712) and three plant extract bioprotectants (AHDB 9957, AHDB 9730 & AHDB 9852). The product application timings differed between products based on information provided on their actual or likely label directions (**Table 1**) as well as the dose rates. Products were applied overhead using a single 02F110 nozzle by air-assisted knapsack sprayer at 400L/ha, with six application timings at weekly intervals. Pots of hebe plants with the leaf spots Septoria and Stemphylium were placed within the trial area the day after the second application day to act as disease spreaders and the trial overhead irrigated.

Table 1. Treatment application timings and the products applied to hebe, commencing on 18 October, and completing on 22 November 2022.

Treat-ment	Timing 1 (Day 0)	Timing 2 (Day 7)	Timing 3 (Day 14)	Timing 4 (Day 21)	Timing 5 (Day 28)	Timing 6 (Day 35)
1	Untreated	Untreated	Untreated	Untreated	Untreated	Untreated
2	-	-	Signum	Signum	-	-
3	-	-	Luna Privilege	Luna Privilege	-	-
4	-	-	AHDB 9714	AHDB 9714	-	-
5	AHDB 9713	AHDB 9713	AHDB 9713	-	-	-
6	AHDB 9712	AHDB 9712	AHDB 9712	AHDB 9712	AHDB 9712	AHDB 9712
7	AHDB 9957	AHDB 9957	AHDB 9957	AHDB 9957	AHDB 9957	AHDB 9957
8	AHDB 9730	AHDB 9730	AHDB 9730	AHDB 9730	AHDB 9730	AHDB 9730
9	AHDB 9852	AHDB 9852	AHDB 9852	AHDB 9852	AHDB 9852	AHDB 9852

The plots were assessed immediately preceding the sprays on 18 October, and then similarly on subsequent application days of 25 October, 1, 8, 15 and 22 November. The final assessment was on 13 December, three weeks after the last applications.

At each of the seven assessments, the leaf area covered by Septoria and Stemphylium lesions was assessed individually on each of four mature leaves selected at random on each of the 15 plants in a plot. This was supplemented by whole plot assessments of the % of leaf area covered by both leaf spots, together with a plant vigour index and a record of any phytotoxicity.

Results

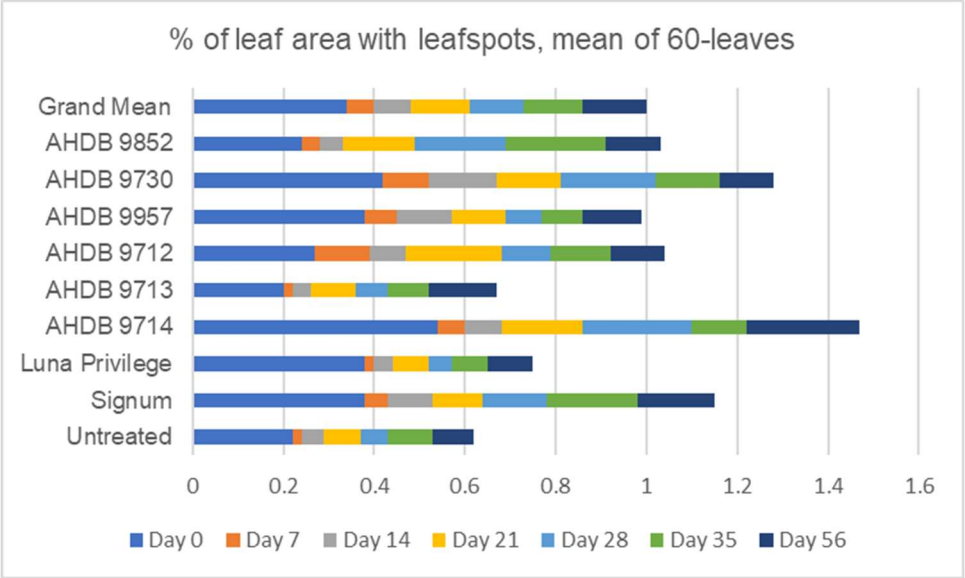

Figure 1. Comparisons of mean % leaf area with leaf-spotting at each of the seven assessment dates based on 60-leaf/plot samples, as shown by each bar segment length scaled on the bottom axis.

There were no significant differences at any of the assessments between the untreated and any of the standard or experimental products in the mean proportion of leaf area affected by the two leafspots based on results from each of 60 leaves per plot (**Figure 1**). Levels following the pre-treatment assessment (Day 0) were mainly below a mean of 0.2% per leaf. The leaves scored were not necessarily the same at each assessment.

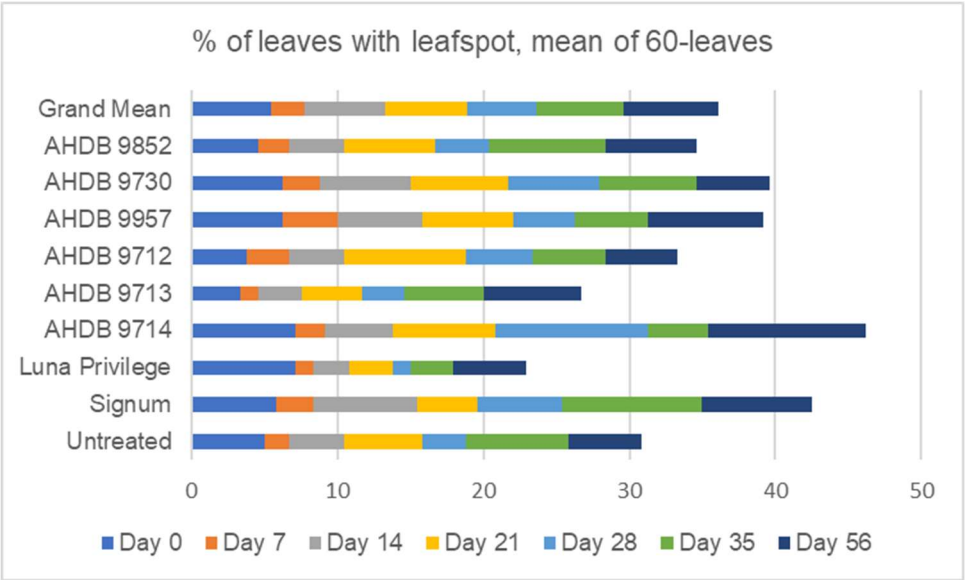

Figure 2. Comparisons of mean % of leaves with leaf-spotting at each of the seven assessment dates based on 60-leaf/plot samples, as shown by each bar segment length scaled on the bottom axis.

There were no significant differences at any of the assessments between the untreated and any of the standard or experimental products in the mean proportion of leaves affected by the two leafspots based on results from each of 60 leaves per plot (**Figure 2**). In general, around 6% of leaves had spotting by one or other, or both Septoria and Stemphylium. The leaves scored were not necessarily the same at each assessment. The experimental products were all applied at least on Days 0, 7 and 14 whereas Luna Privilege and Signum were applied on Days 14 and 21 only. Luna Privilege had the lowest ranked leafspot incidence on Days 28 and 35.

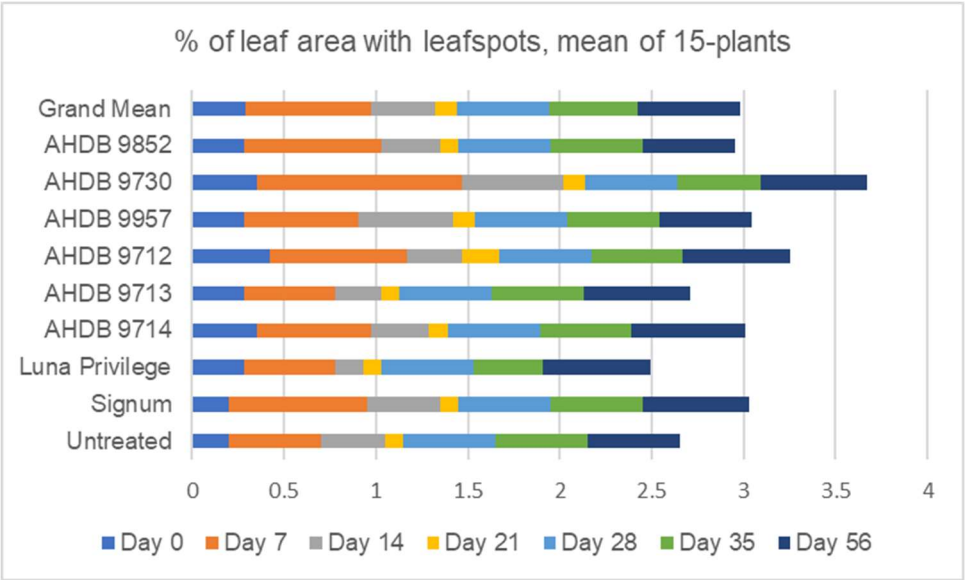

Figure 3. Comparisons of mean % leaf area with leaf-spotting at each of the seven assessment dates based on 15 plants / plot samples, as shown by each bar segment length scaled on the bottom axis.

Assessment across whole plots, rather than on 60 leaves, gave similar results to scores for individual leaves, with at most a mean 0.5% of leaf area with leaf spot

symptoms and no significant differences between the untreated, standard products and experimental products at any of the seven assessments (**Figure 3**).

None of the products caused any phytotoxicity to the hebe plants.

Take home message:

None of the experimental treatments can be recommended for use against fungal leaf spot on hebe as they did not offer any significant reduction compared with the untreated. There were, however, no differences in leaf spotting incidence or severity between the experimental products and the standard conventional products Signum and Luna Privilege. There was little change in symptom severity or incidence over the eight weeks and so there may have been little new infection during the period of the trial to evaluate the control by protectant products.

SCIENCE SECTION

Objectives

To screen plant protection products (chemical, microbial and botanical), for efficacy against fungal leaf spots of hebe and any phytotoxicity.

Methods

The trial was sited at a commercial nursery and set up in a glasshouse on 18 October 2022 with *Hebe rakaiensis* liners from that nursery in 10.5 cm diameter pots. Each plot comprised a plastic carry-tray with drainage holes with three lines of five pots of plants, with about 20 mm space between plants (**Tables 3 & 4**). The layout and a photograph of the trial are given in the Appendix (**Figure 1 & 2**). A randomised block design was used for the trial layout, with nine treatments including an untreated control in each of four replicate columns of trays (**Table 5**).

Trial plants were infected using spreader plants (*Hebe* plants that were naturally infected on the same site), which were introduced on the 8th day after the first spray application and were spread evenly throughout the trial, with one spreader pot in the 300 mm space between each plot within each replicate and at the ends. The spreader plants were kept free from any experimental product applications and remained in-situ until after the final treatment assessments. Following standard nursery practice, the trial received overhead irrigation according to the needs of the plants. A “rain” gauge was placed within the crop to monitor water application.

Prior to the trial, the plants had all received prophylactic treatment by the nursery with a sequence of single applications at seven-day intervals of Previcur Energy (propamocarb) against downy mildew, DiPel DF (*Bacillus thuringiensis* subs. *kurstaki*) against lepidopterous larvae, Amylo-X WG (*Bacillus amyloliquefaciens* subsp. *plantarum* strain D747), and Serenade ASO (*Bacillus amyloliquefaciens* strain QST 713) against fungal diseases. The last nursery application was made on 14 September 2022, a month before experimental product applications commenced.

Five of the treatments were protectant products (AHDB 9713, AHDB 9712, AHDB 9957, AHDB 9730 & AHDB 9852) and so first applied at the start of the trial at Day 0. Curative, as well as protectant products (Signum, Luna Privilege & AHDB 9714) were not applied until Day 14 (**Tables 1 & 6**). The number of subsequent applications was based either on label information or on the probable maximum number of applications likely to be stated for any subsequent Extension of Authorisation for Minor Use (EAMU)

(**Tables 1 & 6**). Dose rates were as on the product label or, for the experimental products, as agreed with the product suppliers (**Table 6**). All treatments were applied at 7-day intervals using a single 02F110 nozzle and an air-assisted knapsack sprayed at 400 L/ha water volume and were sprayed directly above each plant, without run-off (**Table 7**). Due to the density of leaves on hebe and their arrangement down near-vertical stems, good coverage of the lower leaves was not possible. The applied chemicals were mixed directly before spraying (at most 20 minutes before).

The plots were assessed on seven different occasions (**Table 8**), focusing on disease incidence and severity. The assessments were carried out at the time of application of the first treatment, and again at 7-day intervals prior to each treatment application (six applications) and then after three weeks i.e., at 56 days after the first products were applied in the trial.

Two disease assessments were performed: 1) 60-leaf assessments in which four leaves per plant on each of the 15 plants in a plot were assessed and individually recorded for the % cover, separately, of Septoria and Stemphylium leaf spots. This assessment thus also recorded the proportion of leaves affected per plot, and 2) whole plot assessments in which the % of leaf area covered by both leaf spots was assessed across all the pots in a plot. Photographs giving examples of the % cover of each of the leaf spots are given in Appendix **Figures 4 & 5**.

Whole plot vigour was assessed using a 0 (dead) to 9 (excellent) index which considered factors including plant canopy density, the production of new growth and leaf colouration. The plants were examined for any phytotoxicity, for example distortion or stunting. Records were made of any spray deposit residues remaining on the plants.

Trial conduct

Table 2: EPPO guidelines followed, and the variations made for disease assessment.

Relevant EPPO guideline(s)		**Variation from EPPO**
EPPO PP1/135 (4)	Phytotoxicity assessment	No variation
EPPO PP1/152 (4)	Guideline on design and analysis of efficacy evaluation trials	No variation
EPPO PP1/225 (2)	Minimum effective dose	No variation
EPPO PP1/181 (4)	Conduct and reporting of efficacy evaluation trials including good experimental practice	No variation
EPPO PP 1/214 (3)	Principles of acceptable efficacy	No variation
EPPO PP 1/224 (2)	Principles of efficacy evaluation for minor uses	No variation
EPPO PP 1/196 (2)	Efficacy evaluation of fungicides: Fungi on woody ornamentals	From each plot select at random at least 50 leaves of similar age. Record the level of infection: number of infected leaves and percentage of leaf area affected. A scale may be used and, if so, should be described. Variation: 4 mature leaves at random from each of 15 plants/plot given a % leaf spot cover score individually for each disease : % of leaves affected in the sample calculated. Whole plot assessment of % area of any leaf spotting .

EPPO PP 1/221(1)	Efficacy evaluation of fungicides: Foliar diseases on non-woody ornamentals	At least 10 plants or shoots should be selected in each plot. If infection on plants is uneven, at least 5 leaves per plant or shoot should be selected at the position where infection occurs on the untreated plants. The level of infection should be recorded as number of infected leaves and percentage of leaf area infected. A scale may be used and, if so, should be described. If infection on plants is even, it is also possible to assess infection of the whole plant. If plants with very small leaves are used, the level of infection should be estimated for whole shoots. Variation: 4 leaves at random scored individually for each disease separately, plus whole plot % leaf spot cover estimated.

Test site

Table 3: Description of trial location and husbandry

Item	Details
Location address	Wyevale Hereford Kings Acre Road, Hereford, Herefordshire HR4 0SE. Grid Reference SO 47300 42118.
Crop	Hebe
Cultivar	*Hebe rakaiensis*
Soil or substrate type	Peat based growing-media pH 7.0. P 21.2 mg/L (index 2), K 235 mg/L (index 1) Mg 70.9 mg/L (index 4). Organic matter 70.9%. on 14/11/2022
Agronomic practice	Standard glasshouse-grown container crop with overhead (fixed and hosepipe) irrigation. Frost-protection using air blast heating.
Prior history of site	The target diseases have been reported from the nursery.

Trial design

Table 4: Trial layout and number of plants

Item	Details
Trial design:	Randomised block
Number of replicates:	4
Row spacing:	0.3 m between plots & 0.4m between blocks
Plot size: (w x l)	0.31m x 0.56m (pots stood in a 15-hole carry-tray)
Plot size: (m^2)	0.17
Number of plants per plot:	15 in 10.5 cm diameter pots

The plot randomisation and a photograph of the plots are given in the Appendix (**Figures 1 and 2**) together with a photograph of the first plot at the start and finish of the experiment (**Figure 3**).

Treatment details

Table 5: Products, with experimental products coded, together with the quantity of active substance in the product and the product formulation type. Active ingredients are only given for authorised products, otherwise the nature or the substance is given.

AHDB Code	Active substance	Product name/ manufacturer code	Formulation batch number	Content of active substance in product	Formu -lation type	Adju- vant
No need	Boscalid + pyraclostro bin	Signum (MAPP 11450)	12-k00884	267 g/kg and 67 g/kg	WDG	N
No need	Fluopyram	Luna Privilege (MAPP 18393)	EM4L026452	500 g/L	EC	N
AHDB 9714	Confidential					N
AHDB 9713	Confidential					N
AHDB 9712	Confidential					N
AHDB 9957	Confidential					N
AHDB 9730	Confidential					N
AHDB 9852	Confidential					N

Application schedule

Table 6: Treatments, the application rates used. The application slots each product had in the treatment are shown (the application dates are given in **Table 7**).

Treatment number	Treatment product name or AHDB code	Rate of product (l or kg/ha)	Application code
1	Untreated (water sprayed)	n.a.	A,B,C,D,E,F
2	Signum	1.35 kg	C,D
3	Luna Privilege	0.225 L	C,D
4	AHDB 9714	0.35 L	C,D
5	AHDB9713	0.5 kg	A,B,C
6	AHDB9712	0.25 kg	A,B,C,D,E,F
7	AHDB 9957	3.0 L	A,B,C,D,E,F
8	AHDB 9730	1.6 L (0.4% dilution)	A,B,C,D,E,F
9	AHDB 9852	3.0 L	A,B,C,D,E,F

Application details

Table 7: Crop growth, spray equipment and environmental conditions in the glasshouse for each of the weekly application dates.

	Application A	Application B	Application C	Application D	Application E	Application F
Application date	18/10/2022	25/10/2022	1/11/2022	8/11/2022	15/11/2022	22/11/2022
Time of day	12:55	12:42	12:00	12:55	11:26	13:00
Crop growth stage (Max, min average BBCH)	Branching, non-flowering	Branching, non-flowering	Branching, non-flowering	Branching, non-flowering	Branching, non-flowering	Branching, non-flowering
Crop height (cm)	5	5-7	8	7-10	10-15	15
Crop coverage (%)*	N/A	N/A	N/A	N/A	N/A	N/A
Application Method	Foliar	Foliar	Foliar	Foliar	Foliar	Foliar
Application Placement	Spray	Spray	Spray	Spray	Spray	Spray
Application equipment	Oxford sprayer	Oxford sprayer	Oxford sprayer	Oxford sprayer	Oxford sprayer	Oxford sprayer
Nozzle pressure (bar)	3	3	3	3	3	3
Nozzle type	Hypro	Hypro	Hypro	Hypro	Hypro	Hypro
Nozzle size	02F110	02F110	02F110	02F110	02F110	02F110
Application water volume/ha	400	400	400	400	400	400
Temperature of air - shade (°C)	13.2	19.5	13.15	12.85	12.6	9.7
Relative humidity (%)	53.75	77.45	91.38	83.05	53	82.6
Wind speed range (m/s)	n/a	n/a	n/a	n/a	n/a	n/a
Dew presence (Y/N)	N	N	N	N	N	N
Wetness of substrate	Damp	Damp	Damp	Damp	Damp	Damp
Cloud cover (%)	0	75	75	100	100	0

***Foliage ground cover was mostly 100% per plant pot. Foliage closed-up between plots during the trial to leave about 15% of the carry-tray open from above.

Assessment details

Table 8: Evaluation timings following the first of two applications of the two conventional plant protection products on 1 November. All the experimental products were microbial or chemical biopesticides with all but one product (AHDB 9714) first being applied on 18 October (as detailed in **Table 1** &**Table 7**).

	Evaluation Timing (DA)*				
Evaluation date	**After first conventional pesticides**	**After first bio-pesticides**	**Crop Growth Stage (BBCH)**	**Evaluation type (efficacy, phytotoxicity)**	**Assessment**
18/10/2022 (Day 0)	NA	0	Branching, not-flowering	Efficacy	% of leaf area with each leaf spot per leaf from a sample of 60 leaves/plot. % of leaf area covered by leaf spots per plot
				Vigour	0 to 9 (dead to excellent) indices
				Phytotoxicity	% of leaf area affected
25/10/2022 (Day 7)	NA	7	Branching, not-flowering	Efficacy	% of leaf area with each leaf spot per leaf from a sample of 60 leaves/plot. % of leaf area covered by leaf spots per plot
				Vigour	0 to 9 (dead to excellent) indices
				Phytotoxicity	% of leaf area affected
01/11/2022 (Day14)	0	14	Branching, not-flowering	Efficacy	% of leaf area with each leaf spot per leaf from a sample of 60 leaves/plot. % of leaf area covered by leaf spots per plot
				Vigour	0 to 9 (dead to excellent) indices
				Phytotoxicity	% of leaf area affected
08/11/2022 (Day 21)	7	21	Branching, not-flowering	Efficacy	% of leaf area with each leaf spot per leaf from a sample of 60 leaves/plot. % of leaf area covered by leaf spots per plot
				Vigour	0 to 9 (dead to excellent) indices
				Phytotoxicity	% of leaf area affected
15/11/2022 (Day 28)	14	28	Branching, not-flowering	Efficacy	% of leaf area with each leaf spot per leaf from a sample of 60 leaves/plot. % of leaf area covered by leaf spots per plot
				Vigour	0 to 9 (dead to excellent) indices

				Phytotoxicity	% of leaf area affected
			Branching, not-flowering	Efficacy	% of leaf area with each leaf spot per leaf from a sample of 60 leaves/plot. % of leaf area covered by leaf spots per plot
22/11/2022 (Day 35)	21	35	Branching, not-flowering	Efficacy	% of leaf area with each leaf spot per leaf from a sample of 60 leaves/plot. % of leaf area covered by leaf spots per plot
				Vigour	0 to 9 (dead to excellent) indices
				Phytotoxicity	% of leaf area affected
13/12/2022 (Day 56)	28	42	Branching, not-flowering	Efficacy	% of leaf area with each leaf spot per leaf from a sample of 60 leaves/plot. % of leaf area covered by leaf spots per plot
				Vigour	0 to 9 (dead to excellent) indices
				Phytotoxicity	% of leaf area affected

* DA – days after application

The whole-plot disease assessments included new growth that was unlikely to display symptoms of infection until after a latent period (which for the cereal pathogen *Septoria tritici* is 14-28 days ahdb.org.uk/knowledge-library/septoria-tritici-in-winter-wheat). The whole-plot assessments can differ from separate leaf assessments as human bias when sampling leaves can result in selection of affected leaves rather than random sampling of mature leaves. Leaves for the 60-leaf assessments were scored on the plant, the leaves were not detached. Results for the same assessment method were valid for comparison between treatments. All assessments were carried out by the same ADAS staff member following checking of the scoring levels with another staff member.

Statistical analysis

This experiment used a randomised block design and comprised nine treatments, including an untreated control. There were four replicates for each treatment.

Separate means were calculated for Septoria and for Stemphylium from the assessment of % leaf area affected, using four leaves of 15 plants per plot, to make up 60 leaves in total. Both the separate diseases and the combined data for both leaf spots were analysed from the 60-leaf assessments of Septoria and Stemphylium leaf spot using Analysis of Variance. As the same 60 leaves were scored for the % cover of each disease, calculation of the proportion of leaves with each disease, and also with either disease, was carried out and Analysis of Variance performed.

Treatment means were calculated for the % leaf area covered by leaf spots using whole plot assessments and Analysis of Variance calculated.

In addition, the calculated means for both severity (% leaf area) and incidence (number affected out of 60 /plot) were ranked using Duncan's multiple range test to see if there were any products with consistently lower ranking disease levels.

Vigour index scores were compared by Analysis of Variance at the assessment made on the second spray date, but subsequent plot scores were seen to be the same and treatment means alone were presented.

Genstat (21st edition) was used by ADAS Statistician, Chris Dyer.

Results

Phytotoxicity
No phytotoxic effects were seen to follow application of any of the applied treatments during this trial.

Residues
No noticeable spray residues were left on the leaves. AHDB 9852 concentrate was noted as being very viscous but dispersed in the spray tank on dilution with water.

Vigour
Vigour scores are recorded in **Table 9.** Vigour was not very strong at the start but increase over the trial duration. The change in plant vigour is shown in the photograph in the Appendix (**Figure 3**). Plant vigour was the same across all treatments at each assessment. The growth spurt in mid-November followed a period of unusually mild weather for the time of year, with air temperatures at plant height around 14°C. Temperature was not consistently below 10°C until the end of that month. Humidity was usually above 80% throughout the trial period (Appendix **Figure 6** logger chart).

Table 9. Mean vigour scores per plot assessed at seven-day intervals from the day of first treatment applications and finally at 21 days after the last treatment was applied.

Mean plant vigour per plot (0-9, where 9=excellent)							
Treatment	**Day 0**	**Day 7**	**Day 14**	**Day 21**	**Day 28**	**Day 35**	**Day 56**
	18/10/22	25/10/22	01/11/22	08/11/22	15/11/22	22/11/22	13/12/22
Untreated	*	4.5	5	5	6	6	7
Signum	*	5.0	5	5	6	6	7
Luna Privilege	*	5.0	5	5	6	6	7
AHDB 9714	*	5.0	5	5	6	6	7
AHDB 9713	*	4.75	5	5	6	6	7
AHDB 9712	*	5.0	5	5	6	6	7
AHDB 9957	*	4.75	5	5	6	6	7
AHDB 9730	*	4.75	5	5	6	6	7
AHDB 9852	*	5.0	5	5	6	6	7
Grand Mean	*	4.86	5	5	6	6	7
F- Probability value	-	0.461	n.sig.	n.sig.	n.sig.	n.sig.	n.sig.
Least significant difference	-	0.530	0	0	0	0	0
Degrees of freedom	-	24	24	24	24	24	24

n.sig. = scores, and thus mean values, were identical and so ANOVA was not performed.

* Vigour indices were not recorded at trial set-up, but all plants used were of equivalent size.

Disease control

The incidence of infection, of 5%, for the plots that then remained untreated was the same at the start and finish (**Table 10**). The same proportions of leaves were affected initially in the untreated plots by Stemphylium and Septoria, each 2.5%, **(Table 10)**. The larger lesions that develop in Septoria resulted in a marginally greater leaf area cover than Stemphylium (**Table 10**). By the end of the trial the combined incidence of both diseases in the untreated was again 5%, with a slightly greater incidence of Septoria than Stemphylium, however the combined area of leaf spotting was only 0.1% (**Table 10**).

Before the experimental applications commenced, plants in all treatments had a small amount of leaf spotting (**Tables 11 & 12**). At Day 0 across all nine treatments a mean 0.3% of leaf area had leaf spotting, based on both individual leaf assessments (**Table 11**) and whole plot estimations (**Table 13)** with a mean 5.5% of leaves affected based on 60-leaf records (**Table 12**). Leaf spotting at the start could not be avoided as the nursery site and variety were chosen for their predisposition to leaf spotting and because individual hebe leaves are retained between years there was a likelihood of historical leaf spotting.

Table 10 Untreated levels of pathogens (severity and incidence) at application and by the end of the assessment period using the records for 60 leaves examined per plot.

Common name	Scientific Name	EPPO Code	Infestation level pre-application 18/10/2022	Infestation level at assessment period start 18/10/2022	Infestation level at assessment period end 13/12/2022
Stemphylium	*Stemphylium solani*	STEMSO	0.094% leaf area spotted. 2.5% of leaves.	0.094% leaf area spotted. 2.5% of leaves.	0.0125% leaf area spotted. 1.25% of leaves.
Septoria	*Septoria exotica*		0.125% leaf area spotted. 2.5% of leaves.	0.125% leaf area spotted. 2.5% of leaves.	0.079% leaf area spotted. 3.75% of leaves.
Both leaf spots	*S. solani* + *S. exotica*		0.219% leaf area spotted. 5.0% of leaves.	0.219% leaf area spotted. 5.0% of leaves.	0.092% leaf area spotted. 5.0% of leaves.

Following ANOVA statistical analysis of the 60-leaf scores, neither the combined, nor the separately assessed Septoria and Stemphylium disease data were significantly different from the untreated control. No consistent ranking of products was seen from Duncan's tests.

The combined disease severity and incidence from the 60-leaf samples is presented in **Table 11** (mean % leaf area affected) and **Table 12** (mean % of leaves with leafspot). The proportion of leaves with any leaf spotting was mainly in the region of 6% (**Table 12**), which could affect the marketability of the plants even though the proportion of the leaf area affected by both pathogens combined was principally under 0.5% (**Table 11**).

For the whole plot assessments (**Table 13**), the combined mean leaf spotting only exceeded 0.75% once across the treatment means for all seven assessments. The lowest mean % leaf area with leaf spot across treatments was on the fourth assessment, on 8 November, at 0.12% when there had been new leaf growth, but disease symptom severity then increased a little to reach a mean 0.56% of leaf area by 13 December.

The data for the separate disease scores form the 60-plants is not tabulated as the values were small and the data trend is reflected in the combined data. The Grand Mean of untreated and treated at Day 0 for Septoria cover was 0.2%, and Stemphylium 0.1%, and by Day 35 it was 0.1% and 0.02%, respectively. The incidence of Septoria at Day 0 was 3.6% when Stemphylium was 1.9%, and by the assessment at the last spray on Day 35 their incidences were 4.3% and 1.7%, respectively. Isolations made from leaf samples indicated that early spotting by Septoria could be indistinguishable from that of Stemphylium spotting, thus supporting the preferred use of combined data.

Table 11. Mean disease severity per 60 leaves assessed at seven-day intervals from the first treatment applications and finally at 21 days after the last treatment was applied. Mean proportion of leaf area covered by leaf spotting of either disease, on the leaves assessed.

Mean % leaf area with leaf spotting based on 60 leaves examined per plot							
Treatment	**Day 0** 18/10/22	**Day 7** 25/10/22	**Day 14** 01/11/22	**Day 21** 08/11/22	**Day 28** 15/11/22	**Day 35** 22/11/22	**Day 56** 13/12/22
Untreated	0.22	0.02	0.05	0.08	0.06	0.10	0.09
Signum	0.38	0.05	0.10	0.11	0.14	0.20	0.17
Luna Privilege	0.38	0.02	0.04	0.08	0.05	0.08	0.10
AHDB 9714	0.54	0.06	0.08	0.18	0.24	0.12	0.25
AHDB 9713	0.20	0.02	0.04	0.10	0.07	0.09	0.15
AHDB 9712	0.27	0.12	0.08	0.21	0.11	0.13	0.12
AHDB 9957	0.38	0.07	0.12	0.12	0.08	0.09	0.13
AHDB 9730	0.42	0.10	0.15	0.14	0.21	0.14	0.12
AHDB 9852	0.24	0.04	0.05	0.16	0.20	0.22	0.12
Grand Mean	0.34	0.06	0.08	0.13	0.12	0.13	0.14
F- Probability value	0.709	0.477	0.378	0.244	0.487	0.66	0.82
Least significant difference	0.402	0.107	0.107	0.114	0.198	0.175	0.189
Degrees of freedom	24	24	24	24	24	24	24

Day 7 to Day 14 scores for Signum, Luna Privilege and AHDB 9714 precede treatment, other products were applied directly following Day 0 assessment.

Table 12. Mean disease incidence per 60 leaves assessed at seven-day intervals and finally at 21 days after the last treatment was applied. Mean proportion (%) of the leaves assessed that had leaf spotting by either disease. Day 0 was before any spray applications.

Mean % of 60 leaves examined per plot which had either Septoria or Stemphylium							
Treatment	**Day 0** 18/10/22	**Day 7** 25/10/22	**Day 14** 01/11/22	**Day 21** 08/11/22	**Day 28** 15/11/22	**Day 35** 22/11/22	**Day 56** 13/12/22
Untreated	5.00	1.67	3.75	5.42	2.92	7.08	5.00
Signum	5.83	2.50	7.08	4.17	5.83	9.58	7.50
Luna Privilege	7.08	1.25	2.5	2.92	1.25	2.92	5.00
AHDB 9714	7.08	2.08	4.58	7.08	10.42	4.17	10.83
AHDB 9713	3.33	1.25	2.92	4.17	2.92	5.42	6.67
AHDB 9712	3.75	2.92	3.75	8.33	4.58	5.00	5.00
AHDB 9957	6.25	3.75	5.83	6.25	4.17	5.00	7.92
AHDB 9730	6.25	2.50	6.25	6.67	6.25	6.67	5.00
AHDB 9852	4.58	2.08	3.75	6.25	3.75	7.92	6.25
Grand Mean	5.46	2.22	5.54	5.69	4.68	5.97	6.57
F- Probability value	0.752	0.890	0.700	0.441	0.149	0.497	0.506
Least significant difference	5.086	3.582	4.490	4.867	5.908	6.117	5.922
Degrees of freedom	24	24	24	24	24	24	24

Day 7 & Day 14 scores for Signum, Luna Privilege and AHDB 9714 precede treatment, other products were applied directly following Day 0 assessment (see **Tables 1** & **6** for details)

Table 13. Whole plot assessments of percentage of leaf area with leaf spotting by either disease estimated to obtain treatment means. Assessed at seven-day intervals from the first treatment applications and finally at 21 days after the last treatment was applied. Day 0 was before any spray applications.

Mean % leaf area with leaf spotting per plot across all leaves							
Treatment	**Day 0** 18/10/22	**Day 7** 25/10/22	**Day 14** 01/11/22	**Day 21** 08/11/22	**Day 28** 15/11/22	**Day 35** 22/11/22	**Day 56** 13/12/22
Untreated	0.20	0.50	0.35	0.10	0.50	0.50	0.50
Signum	0.20	0.75	0.40	0.10	0.50	0.50	0.58
Luna Privilege	0.28	0.50	0.15	0.10	0.50	0.38	0.58
AHDB 9714	0.35	0.62	0.32	0.10	0.50	0.50	0.62
AHDB 9713	0.28	0.50	0.25	0.10	0.50	0.50	0.58
AHDB 9712	0.42	0.75	0.30	0.20	0.50	0.50	0.58
AHDB 9957	0.28	0.62	0.52	0.12	0.50	0.50	0.50
AHDB 9730	0.35	1.12	0.55	0.12	0.50	0.45	0.58
AHDB 9852	0.28	0.75	0.32	0.10	0.50	0.50	0.50
Grand Mean	0.29	0.68	0.35	0.12	0.50	0.48	0.56

F- Probability value	0.58	0.101	0.37	0.577	n.sig	0.955	0.906
Least significant difference	0.233	0.417	0.343	0.105	0.000	0.225	0.205
Degrees of freedom	24	24	24	24	24	24	24

Day 7 to Day 14 scores for Signum, Luna Privilege and AHDB 9714 precede treatment, other products were applied directly following Day 0 assessment.

Discussion

None of the applied treatments offered any statistically significant reduction in either of the two leaf spot diseases. The symptoms that were present at a low level at the start of the experiment would not be expected to be eradicated by any product. Although conditions were suitable for disease development (warm and humid, with water splash for spread from spreader pots and between closely spaced foliage) there was negligible symptom development over the period of the trial even in the untreated. Because hebe leaves are evergreen, any nursery application of a curative fungicide in early 2022 could have reduced pathogen mycelium viability within the visible spots and so even though conditions were conducive for infection there may have been little or no spore production.

It was noted that the plant extract AHDB 9852 concentrate was an extremely viscous liquid and so it could be difficult to measure out accurately when relatively small volumes are required to treat limited areas on a nursery.

Conclusions

Based on the outcomes of this trial, none of the included treatments can be recommended for use against fungal leafspot on hebe. No phytotoxic effects, nor unacceptable spray deposits, were recorded during the trial and so all the experimental products would be suitable for any further efficacy testing on hebe.

Acknowledgements

ADAS is grateful to the host nursery for being willing to participate in this project and for the supply of plants, providing a glasshouse area and watering. Also, the co-operation of plant protection company representatives for providing information on their products and supplying samples for testing.

Appendix

Trial assessments & sprays conducted by ADAS Rosemaund staff led by Gabriella Parcell.

Date	Trial Diary
17/08/2022	Nursery application of Previcur Energy downy mildew fungicide
31/08/2022	Nursery application of DipPel DF insecticide
07/09/2022	Nursery application of Amylo X protectant fungicide
14/09/2022	Nursery application of Serenade ASO protectant fungicide
18/10/2022	Trial set-up on a Tuesday with plants of similar vigour in all plots. Spray and assessment 1 complete. Agreed with nursery that if watering is needed it will be done on Mondays. Trt 9 AHDB 9852 concentrate is very thick and hard to accurately measure using a syringe.
25/10/2022	Spray and assessment 2 complete. 0.5mm of water in the rain gauges.
26/10/2022	Spreader plants put out in trial
01/11/2022	Spray and assessment 3 complete. Rain gauges: 1.7, 3.7, 5 and 3mm. Spreader plants removed before spray and returned afterwards.
08/11/2022	Spray and assessment 4 complete. No water in rain gauges.
15/11/2022	Spray and assessment 5 complete. No water in rain gauges as watered via spot spray method. Lots of new growth on plants this week.
22/11/2022	Spray and assessment 6 complete. No water in rain gauges. Requested change from hosepipe to overhead irrigation using irrigation headers.
13/12/2022	Final assessment done. Data logger removed and downloaded. 4mm and 7.5mm in rain gauges from overhead irrigation. Plants have put on some new growth in last couple of weeks and are looking more vigorous. Three hours reported spent irrigating by the nursery staff over the trial period.

Assessments and Application record raw data source on ADAS Boxworth Local Area Network

\\bw\DATA\data\HORTIC\1022247 - RM-23-001 - AHDB Fungal leaf spot efficacy trial HNS\RM23-001\Data file - SHE plan/RM23-001 HNS Leaf Spot Fungal – Datafile 01.xls

Photographs for each assessment date are on the ADAS Rosemaund Local Area Network

\\RM\data\Arable 2023\Horticulture\RM23-001 HNS Leaf Spot Fungal

TREATMENT	6	8	4	1	7	2	3	9	5
BLOCK	4	4	4	4	4	4	4	4	4
PLOT	401	402	403	404	405	406	407	408	409
TREATMENT	4	6	2	8	7	5	1	3	9
BLOCK	3	3	3	3	3	3	3	3	3
PLOT	301	302	303	304	305	306	307	308	309
TREATMENT	1	3	5	2	6	7	9	4	8
BLOCK	2	2	2	2	2	2	2	2	2
PLOT	201	202	203	204	205	206	207	208	209
TREATMENT	4	7	9	1	3	6	2	5	8
BLOCK	1	1	1	1	1	1	1	1	1
PLOT	101	102	103	104	105	106	107	108	109

Figure 1. Trial design: Layout of hebe plots in the glasshouse, with 15 plants per plot randomised within each of four lines of carry-trays. October to December 2022

Figure 2. Hebe plots in the glasshouse, with 15 plants per plot randomised within each of four lines of nine carry-trays on 13 December 2022. The data logger was held in the terracotta-coloured screen, and the “rain”-gauge was positioned further down the replicate. Note the condensation on the windows which indicates humid conditions favourable to fungal infections.

Plot (101) of 15 hebe plants on 18 October Plot (101) of 15 hebe plants on 13 December

Figure 3. The same plot of hebe liners in their carry-tray at the first and last assessments in 2022, with new leaves grown. Disease spreader pots visible at the head and foot of the plot in the right-hand picture, that were present from the 8th day.

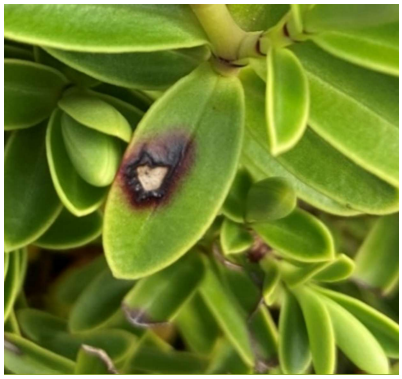

Figure 4. Septoria 20% of a single leaf's area (one of 60 leaves assessed per plot)

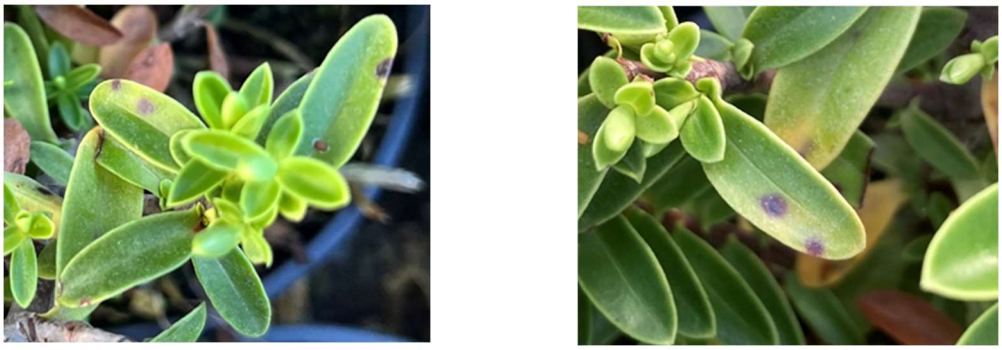

Figure 5. Stemphylium: Left-hand photograph shows 5% of a single leaf's area (one of 60 leaves assessed per plot) and the right-hand photograph shows 2%.

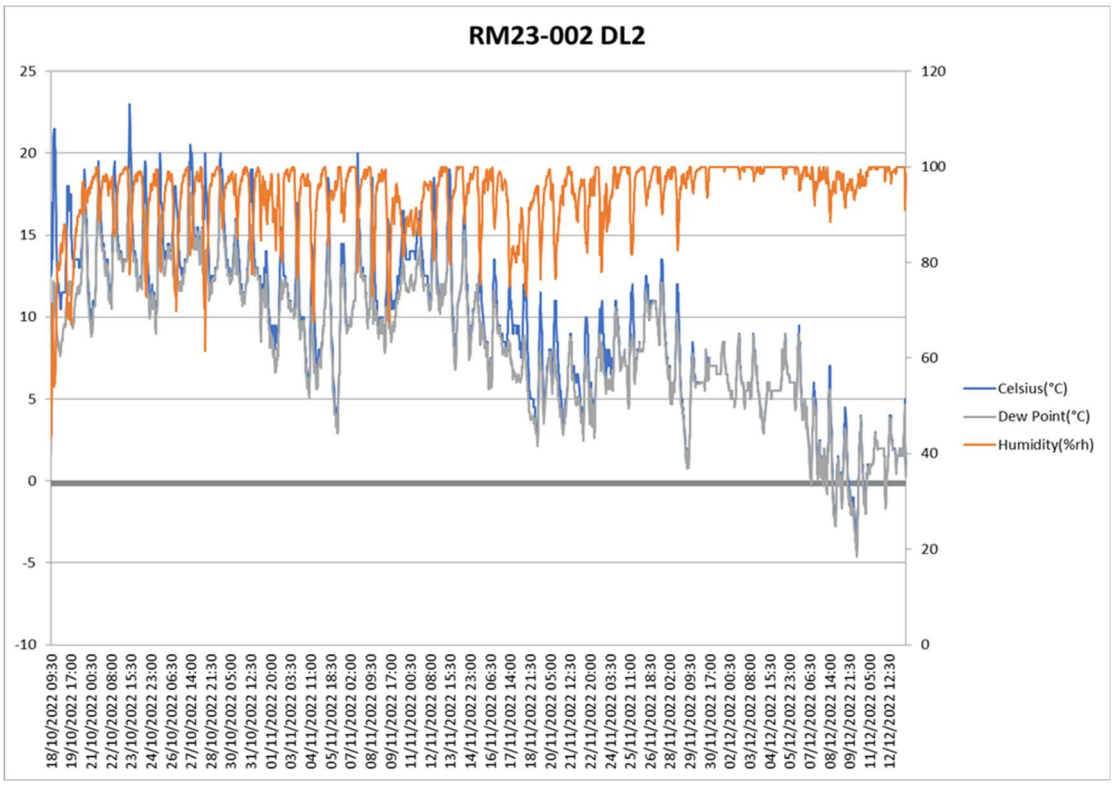

Figure 6. Data logger RM23-002 half-hourly records of air temperature (left-hand axis and blue lines) and humidity (right-hand axis and upper, orange lines) from within a ventilated screen in the nursery glasshouse next to a hebe plot at plant canopy level. Recorded from set up on 18 October 2022 to final assessment on the 13 December. Dew point = calculated times when high humidity and surface temperature would have been conducive to condensation forming on the leaves.

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

Signature Alison Richardson
Authorised signatory

Certification Number
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