

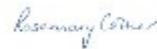
SCEPTREPLUS

Final Trial Report

Trial code:	2017 SP03
Title:	Control of asparagus beetle adults and larvae with novel insecticide and bioinsecticide sprays
Crop	Group: Field vegetables - asparagus
Target	Asparagus beetle – <i>Crioceris asparagi</i>
Lead researcher:	Dr Rosemary Collier
Organisation:	University of Warwick, School of Life Sciences, Wellesbourne, Warwick CV35 9EF
Period:	June 2017 – June 2018
Report date:	24/10/18
Report author:	Andrew Jukes
ORETO Number: (certificate should be attached)	381

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.

12 November 2018



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Date

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Authors signature

Trial Summary

Introduction

The quality of asparagus crops can be reduced by eggs laid by asparagus beetle (*Crioceris asparagi*) on emerging spears at harvest. Also, when the asparagus is allowed to develop into ferns, feeding by asparagus adults and particularly larvae can reduce the vigour of the plants in the following season. There are currently a limited number of control options, none with a sufficiently short harvest interval for treatment of spears, and an over-reliance on a single active/mode of action, which could lead to resistance in the pest population.

Methods

With the kind permission of Philip Langley (G's Fresh) asparagus beetles and plant material were collected from an asparagus field in Tiddington, Warwickshire in both 2017 and 2018. The trial consisted of twelve treatments which were replicated in time, as insects were collected from the field, to give a total of 5 replicates for each insect life stage. The treatments consisted of conventional insecticides and bio-insecticides. Pieces of asparagus fern were kept fresh by inserting them into a block of Oasis® which was kept wet. For trials on larvae the fern was infested with 10 larvae/block of fern before spraying and each block was placed in a separate cage (Bugdorm®, 47.5 x 47.5 x 47.5 cm), and for trials on adults the beetles were released into the cage containing the block of fern (10/cage) after spraying. In both cases the caged insects and blocks of fern were kept at 20°C in the Insect Rearing Unit (IRU) at Wellesbourne. All treatments were applied using a knapsack sprayer fitted with 02F110 nozzles in 300l/ha water. The numbers of live and dead insects were counted 1, 4 and 6 days after spraying.

Results

The mean percentage dead adult and larval asparagus beetles 1, 4 and 6 days after spraying are presented in the table below (Angular transformation was used prior to data analysis but only back-transformed data are presented).

Day	Mean % dead larvae			Mean % dead adults		
	1	4	6	1	4	6
Treatment *bio-insecticide						
Untreated	9.3	28.4	35.8	0	0.41	1.65
Cypermethrin	68.3	93.9	93.9	0.75	5.55	6.10
Benevia	90.5	100	100	1.34	43.58	60.17
AHDB9966	91.1	98.3	99.6	5.87	20.55	23.21
Spruzit*	54.4	89.0	96.8	0	6.11	17.38
AHDB9964*	36.5	81.8	95.7	0	1.65	4.82
AHDB9965	99.6	100	100	33.05	53.89	60.17
AHDB9969	98.7	100	100	20.86	73.23	84.11
Tracer	97.6	98.4	100	5.87	51.04	83.40
Steward	82.6	100	100	3.22	55.15	65.45
AHDB9968*	12.6	88.2	89.5	0	0.41	2.45
AHDB9967*	28.9	81.0	81.0	0	0.41	1.65
	Not significantly different from untreated control (p>0.05)					
	Significantly different from untreated control (p<0.05)					

Conclusions

All of the treatments controlled larvae effectively. Conventional treatments tended to act more quickly than the bio-insecticides and kill a greater proportion of the larvae but all treatments increased mortality to greater than 80%. With no direct contact

action (which would also probably be the case with field applications as the adult beetles are so mobile) the adults were more difficult to kill. Two treatments (ADDB9965 and AHDB9969) increased mortality after 1 day compared with the untreated control and a further 4 treatments (Benevia, AHDB9966, Tracer and Steward) had increased mortality after 4 days. None of the bio-insecticides had significantly increased mortality after 6 days. The most effective treatments after 6 days (Tracer and AHDB9969) increased mortality to greater than 80%.

Take home message:

All of the test products were effective against larvae with only small differences between conventional insecticides and bio-insecticides. Adults were more difficult to kill and all of the bio-insecticides and the standard, cypermethrin, were largely ineffective. Of the conventional insecticides, initial mortality was greater with AHDB9965 and AHDB9969, but after 6 days there was no statistically significant difference between these two products and Benevia, Tracer and Steward.

Objectives

1. To evaluate the effectiveness of conventional and bio-insecticides as foliar sprays for the control of adult and larval asparagus beetles on asparagus
2. To monitor the treated crop 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
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	University of Warwick Wellesbourne Campus Wellesbourne Warwick CV35 9EF
Crop	Asparagus
Cultivar	Unknown
Soil or substrate type	Oasis®
Agronomic practice	Not relevant
Prior history of site	Not relevant

Trial design

Item	Details
Trial design:	Randomized in time
Number of replicates:	5
Row spacing:	Not relevant
Plot size: (w x l)	1 cage (47.5 x 47.5 x 47.5 cm)
Plot size: (m ²)	Not relevant
Number of plants per plot:	1
<i>Leaf Wall Area calculations</i>	Not relevant

Treatment details

AHDB Code	Active substance	Product name/ manufacturers code	Formulation batch number	Content of active substance in product	Formulation type	Adjuvant
	Untreated control					
Authorised	Cypermethrin (standard)	Cypermethrin 500EC	15209600	500 g/l	EC	None
AHDB9948	N/D	N/D	N/D	N/D	N/D	None
AHDB9966	N/D	N/D	N/D	N/D	N/D	None
Authorised (post-harvest)	Pyrethrins	Spruzit	5736/Apr16	4.59g/l	EC	None
AHDB9964	N/D	N/D	N/D	N/D	N/D	None
AHDB9965	N/D	N/D	N/D	N/D	N/D	None
AHDB9969	N/D	N/D	N/D	N/D	N/D	None
Authorised (post-harvest)	Spinosad	Tracer	F055G5Q048	480 g/l	SC	None
Not authorised	Indoxacarb	Steward	FEB16CE172	30%	WG	None
AHDB9968	N/D	N/D	N/D	N/D	N/D	None
AHDB9967	N/D	N/D	N/D	N/D	N/D	None
AHDB9888 ¹	N/D	N/D	N/D	N/D	N/D	None

¹ Additional treatment tested after original trial had concluded

Methods, assessments and records

Application schedule

Treatment number	Treatment: product name or AHDB code (*bio-insecticide)	Rate of active substance (ml or g a.s./ha)	Rate of product (l or kg/ha)	Application code
1	Control			
2	Cypermethrin 500EC	25	0.05	A
3	AHDB9948	75	0.75	A
4	AHDB9966	24	0.2	A
5	Spruzit*	27.5	6	A
6	AHDB9964*	837.5	5	A
7	AHDB9965	75	0.375	A
8	AHDB9969	75	0.3	A
9	Tracer	96	0.2	A
10	Steward	25.5	0.085	A
11	AHDB9968*	15	1.5	A
12	AHDB9967*	144	2.4	A
13	AHDB9888 ¹	60	0.2	A

¹ Additional treatment tested after original trial had concluded

Application details

	Application A1 (1 x larvae)	Application A2 (1 x larvae, 1 x adult)	Application A3 (1 x larvae, 1 x adult)	Application A4 (1 x adult)
Application date	13/7/17	20/7/17	27/7/17	10/8/17
Time of day	13.30	13.30	13.30	13.30
Crop growth stage (Max, min average BBCH)	39 (in field)			
Crop height (cm)	30 (cage height)			
Crop coverage (%)	Not relevant			
Application Method	Spray			
Application Placement	Foliar			
Application equipment	Berthoud Vermorel 2000HP			
Nozzle pressure	2 bar			
Nozzle type	02F110			
Nozzle size	02			
Application water volume/ha	300			
Temperature of air - shade (°C)	Not relevant			
Relative humidity (%)	Not relevant			
Wind speed range (m/s)	Not relevant			
Dew presence (Y/N)	Not relevant			
Temperature of soil - 2-5 cm (°C)	Not relevant			
Wetness of soil - 2-5 cm	Not relevant			
Cloud cover (%)	Not relevant			

	Application A5 (1 x adult)	Application A6 (1 x larvae, 1 x adult)	Application A7 (AHDB9888 - adult)	Application A8 (AHDB9888 - larvae)
Application date	7/9/17	19/9/18	9/8/18	21/8/18
Time of day	13.30	13.30	13.30	13.30
Crop growth stage (Max, min average BECH)	39 (in field)			
Crop height (cm)	30 (cage height)			
Crop coverage (%)	Not relevant			
Application Method	Spray			
Application Placement	Foliar			
Application equipment	Berthoud Vermorel 2000HP			
Nozzle pressure	2 bar			
Nozzle type	02F110			
Nozzle size	02			
Application water volume/ha	300			
Temperature of air - shade (°C)	Not relevant			
Relative humidity (%)	Not relevant			
Wind speed range (m/s)	Not relevant			
Dew presence (Y/N)	Not relevant			
Temperature of soil - 2-5 cm (°C)	Not relevant			
Wetness of soil - 2-5 cm	Not relevant			
Cloud cover (%)	Not relevant			

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
Asparagus beetle	<i>Crioceris asparagi</i>		10 larvae or 10 adults	Not relevant – artificial inoculation	

Assessment details

With the kind permission of Philip Langley (G's Fresh) asparagus beetles and plant material were collected from an asparagus field in Tiddington, Warwickshire in both 2017 and 2018. The trial consisted of twelve treatments which were replicated in time, as insects were collected from the field, to give a total of 5 replicates for each insect life stage. Pieces of asparagus fern were kept fresh by inserting them into a block of Oasis which was kept wet. For trials on larvae the pieces of fern were inoculated with 10 larvae/block of fern before spraying and for adult trials the adults were released into the cages containing blocks of fern (10/cage) after spraying. In both cases the caged ferns were kept at 20°C in the Insect Rearing Unit (IRU) at Wellesbourne. The numbers of live, sick (adults only) and dead insects were counted 1, 4 and 6 days after spraying. Phytotoxicity was assessed 4 days after spraying. Photographs of the set-up are in the Appendix.

Evaluation date	Evaluation Timing (DA)*	Crop Growth Stage (BBCH)	Evaluation type (efficacy, phytotox)	Assessment
14/7/18, 21/7/18, 28/7/18, 11/8/17, 8/9/17, 20/7/18, 9/8/18, 21/8/18	1	39	Efficacy	Number of live and dead adults or larvae
17/7/18, 24/7/17, 31/7/17, 14/8/17, 11/9/17, 23/7/18, 14/8/18, 26/8/18	4	39	Efficacy Phytotoxicity	Number of live and dead adults or larvae. Leaf damage
19/7/17, 26/7/17, 2/8/17, 16/8/17, 13/9/17, 25/7/18, 16/8/18, 28/8/18	6	39	Efficacy	Number of live and dead adults or larvae.

* DA – days after application

Statistical analysis

The trial was analysed by Andrew Mead at Rothamsted Research as a randomised complete block design with 5 replicates (occasions) of 12 treatments, using angular transformed percentages within ANOVA using the Genstat program. No statistical analyses were performed on data from the additional treatment (AHDB9888) tested after the original trial had concluded.

Results

Phytotoxicity

There was no evidence of phytotoxic effects with any treatment.

Asparagus beetle mortality

The results for the cumulative mean percentage dead larvae, dead adults and dead plus sick adults on the 3 occasions after treatment are presented in Table 1 and Figure 1, Table 2 and Figure 2 and Table 3 and Figure 3 respectively. Results significantly different from the untreated control are highlighted in the tables. Table 4 and Figure 4 summarise the results from application of the additional treatment.

Table 1 Cumulative mean percentage dead asparagus beetle larvae 1, 4 and 6 days after spraying (*bio-insecticide).

Day	1		4		6	
	Ang	Back-trans	Ang	Back-trans	Ang	Back-trans
Treatment						
Untreated	17.7	9.3	32.2	28.4	36.8	35.8
Cypermethrin	55.8	68.3	75.7	93.9	75.7	93.9
Benevia	72.1	90.5	90.0	100	90.0	100
AHDB9966	72.6	91.1	82.4	98.3	86.3	99.6
Spruzit*	47.5	54.4	70.7	89.0	79.7	96.8
AHDB9964*	37.2	36.5	64.8	81.8	78.0	95.7
AHDB9965	86.3	99.6	90.0	100	90.0	100
AHDB9969	83.4	98.7	90.0	100	90.0	100
Tracer	81.0	97.6	82.6	98.4	90.0	100
Steward	65.4	82.6	90.0	100	90.0	100
AHDB9968*	20.8	12.6	69.9	88.2	71.1	89.5
AHDB9967*	32.5	28.9	64.2	81.0	64.2	81.0
F value	10.61		9.45		8.89	
P -value	<0.001		<0.001		<0.001	
d.f.	44		44		44	
s.e.d.	10.63		7.74		7.48	
l.s.d.	21.43		15.60		15.07	

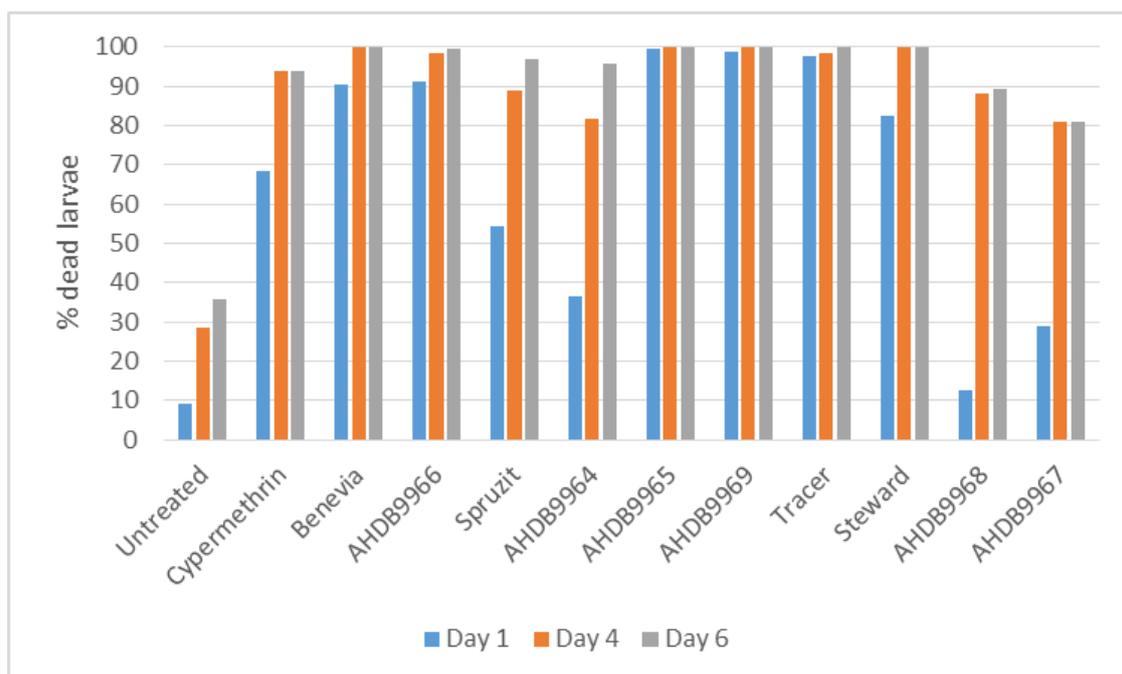


Figure 1 Cumulative mean percentage dead asparagus beetle larvae 1, 4 and 6 days after spraying (Spruzit, AHDB9964, AHDB9968 and AHDB9967 are bio-insecticides).

Table 2 Cumulative mean percentage dead asparagus beetle adults 1, 4 and 6 days after spraying (*bio-insecticide).

Day	1		4		6	
	Ang	Back-trans	Ang	Back-trans	Ang	Back-trans
Treatment						
Untreated	0.0	0	3.7	0.41	7.4	1.65
Cypermethrin	5.0	0.8	13.6	5.55	14.3	6.10
Benevia	6.6	1.3	41.3	43.58	50.9	60.17
AHDB9966	14.0	5.9	27.0	20.55	28.8	23.21
Spruzit*	0.0	0	14.3	6.11	24.6	17.38
AHDB9964*	0.0	0	7.4	1.65	12.7	4.82
AHDB9965	35.1	33.1	47.2	53.89	50.9	60.17
AHDB9969	27.2	20.9	58.8	73.23	66.5	84.11
Tracer	14.0	5.9	45.6	51.04	66.0	83.40
Steward	10.3	3.2	48.0	55.15	54.0	65.45
AHDB9968*	0.0	0	3.7	0.41	9.0	2.45
AHDB9967*	0.0	0	3.7	0.41	7.4	1.65
F value	5.40		9.34		11.67	
P -value	<0.001		<0.001		<0.001	
d.f.	43		43		43	
s.e.d.	7.08		9.63		9.69	
l.s.d.	14.28		19.42		19.54	

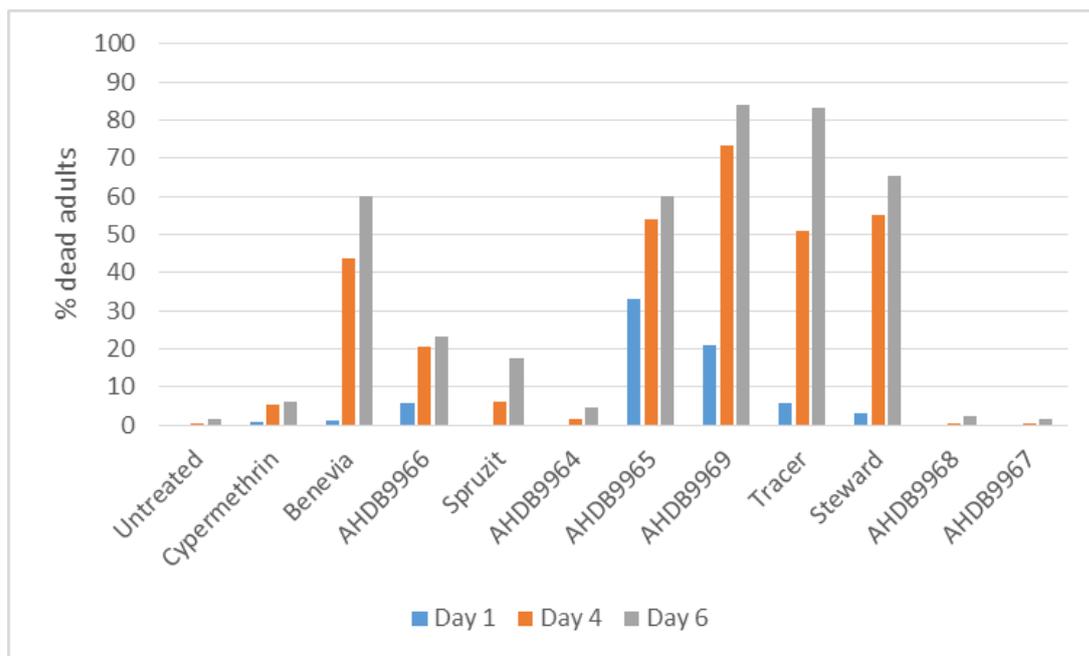


Figure 2 Cumulative mean percentage dead asparagus beetle adults 1, 4 and 6 days after spraying (Spruzit, AHDB9964, AHDB9968 and AHDB9967 are bio-insecticides).

Table 3 Cumulative mean percentage dead or sick asparagus beetle adults 1, 4 and 6 days after spraying (*bio-insecticide).

Day	1		4		6	
	Ang	Back-trans	Ang	Back-trans	Ang	Back-trans
Treatment						
Untreated	0.0	0	3.7	0.4	7.4	1.7
Cypermethrin	8.2	2.0	14.3	6.1	14.0	5.9
Benevia	22.3	14.4	52.1	62.4	58.8	73.2
AHDB9966	15.6	7.3	32.5	28.9	32.7	29.1
Spruzit*	17.0	8.5	24.6	17.4	29.7	24.5
AHDB9964*	3.7	0.4	10.3	3.2	18.0	9.6
AHDB9965	57.7	71.4	61.4	77.1	52.2	62.4
AHDB9969	44.5	49.2	69.5	87.8	75.5	93.7
Tracer	40.9	42.9	59.3	74.0	68.3	86.3
Steward	27.1	20.8	61.8	77.7	64.2	81.1
AHDB9968*	0.0	0	3.7	0.4	9.0	2.5
AHDB9967*	14.0	5.9	9.0	2.5	7.4	1.7
F value	7.40		14.10		13.47	
P -value	<0.001		<0.001		<0.001	
d.f.	43		43		43	
s.e.d.	9.63		9.67		10.00	
l.s.d.	19.42		19.50		20.17	

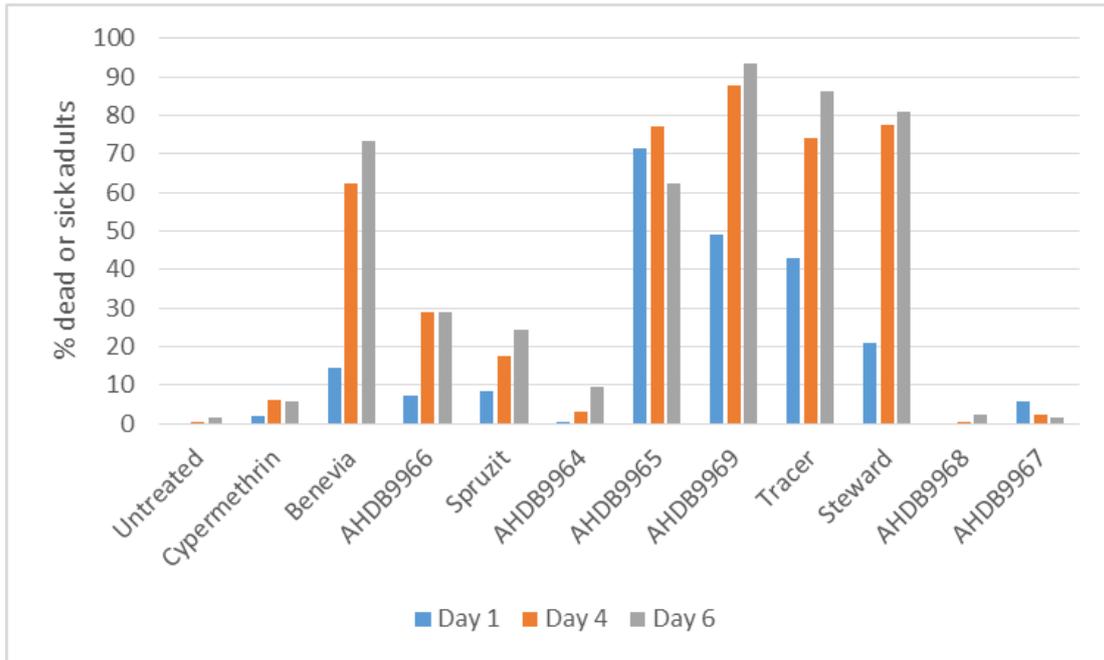


Figure 3 Cumulative mean percentage dead or sick asparagus beetle adults 1, 4 and 6 days after spraying (Spruzit, AHDB9964, AHDB9968 and AHDB9967 are bio-insecticides).

Table 4 Additional treatment - cumulative mean percentage dead, dead or sick asparagus beetle adults and dead asparagus beetle larvae 1, 4 and 6 days after spraying.

Day	1			4			6		
	Adult		Larvae	Adult		Larvae	Adult		Larvae
Assay	% dead	% dead or sick	% dead	% dead	% dead or sick	% dead	% dead or sick	% dead	
Treatment									
Untreated	0	2.5	2.5	0	2.5	2.5	0	2.5	5
AHDB9888	0	2.5	2.5	30	12.5	2.5	86.7	89.4	89.4

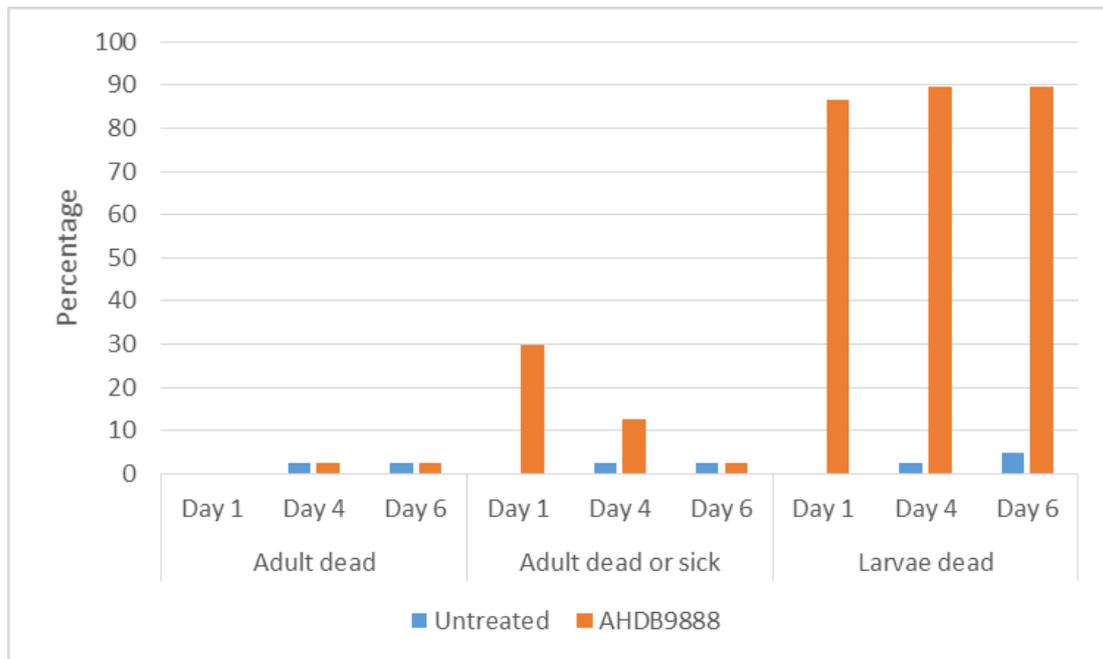


Figure 4 Additional treatment - cumulative mean percentage dead, dead or sick asparagus beetle adults and dead asparagus beetle larvae 1, 4 and 6 days after spraying.

Discussion

All of the treatments increased larval mortality significantly ($p < 0.05$) compared with the untreated control. Conventional treatments tended to act more quickly than the bio-insecticides and kill a greater proportion of the larvae. Six days after treatment all treatments had increased mortality to greater than 80% but there was little difference between the most effective treatments. Only AHDB9967 and AHDB9968 were significantly ($p < 0.05$) less effective. With no direct contact action (as would probably be the case with field applications) the adults were more difficult to kill. Two treatments (AHDB9965 and AHDB9969) had significantly ($p < 0.05$) increased mortality after 1 day and a further 4 treatments (Benevia, AHDB9966, Tracer and Steward) had significantly ($p < 0.05$) increased mortality after 4 days compared with the untreated control. After 6 days AHDB 9966 was significantly ($p < 0.05$) less effective than the other 5 effective treatments. None of the bio-insecticides and the standard cypermethrin treatment had significantly ($p < 0.05$) increased mortality compared with the untreated control after 6 days. The most effective treatments after 6 days (Tracer and AHDB9969) increased mortality to greater than 80%.

The additional treatment (AHDB9888) tested after the main trial controlled larvae effectively but was largely ineffective against adults. Some adults identified as being sick 1 day after spraying subsequently recovered and after 6 days there was virtually no adult mortality.

All treatments mixed and sprayed well. No wetter was required. There were no phytotoxic effects.

Conclusions

- Larvae were easier to kill than adults.
- All treatments increased larval mortality but AHDB9967 and AHDB9968 were less effective.
- All conventional insecticides (except the standard cypermethrin and AHDB9888) increased adult mortality. AHDB9965, AHDB9969, Benevia, Tracer and Steward were the most effective.
- No treatments caused phytotoxic effects.

Acknowledgements

We would like to thank AHDB and the participating crop protection companies for project funding. We would also like to thank Phil Langley of G's Fresh for technical advice and access to an asparagus crop.

Appendix

a. Crop diary – events related to growing crop

Crop	Cultivar	Planting/sowing date	Row width (m)
Asparagus	Not known	Perennial crop	Not relevant

Previous cropping

Year	Crop
2015	Asparagus
2016	Asparagus

Cultivations

Date	Description	Depth
None		

b. Raw data from assessments

Percentage dead larvae (Main trial)

Set up	13/07/2017	20/07/2017	27/07/2017	19/07/2018	19/07/2018
Treatment	Day 1				
1	50	70	80	70	70
2	89	70	100	90	90
3	100	67	70	90	100
4	80	80	50	20	40
5	50	0	10	90	60
6	100	90	100	100	100
7	100	100	100	70	100
8	90	100	80	100	100
9	80	50	100	90	70
10	20	0	60	20	0
11	90	0	10	30	40
12	0	10	10	15	23
	Day 4				
1	100	100	80	90	80
2	100	100	100	100	100
3	100	89	90	100	100
4	90	100	70	80	90
5	70	70	100	80	70
6	100	100	100	100	100
7	100	100	100	100	100
8	90	100	90	100	100
9	100	100	100	100	100
10	90	50	90	100	90
11	100	30	60	70	100
12	10	20	40	38	38
	Day 6				
1	100	100	80	90	80
2	100	100	100	100	100
3	100	100	90	100	100
4	100	100	70	100	90
5	100	80	100	100	70
6	100	100	100	100	100
7	100	100	100	100	100
8	100	100	100	100	100
9	100	100	100	100	100
10	90	60	90	100	90
11	100	30	60	70	100
12	10	50	40	46	38

Percentage dead larvae (AHDB9888)

Set up	09/08/2018	09/08/2018	09/08/2018	09/08/2018
Treatment	Day 1			
1	0	0	0	0
13	80	89	78	100
	Day 4			
1	0	0	0	10
13	80	89	89	100
	Day 6			
1	0	10	0	10
13	80	89	89	100

Percentage dead and dead or sick adults (AHDB9888)

Set up	20/07/2017		27/07/2017		10/08/2017		07/09/2017	
	Dead	Sick/ Dead	Dead	Sick/ Dead	Dead	Sick/ Dead	Dead	Sick/ Dead
Treatment	Day 1							
1	0	0	0	0	0	0	0	0
13	0	10	0	40	0	10	0	60
	Day 4							
1	0	0	10	10	0	0	0	0
13	0	10	10	30	0	10	0	0
	Day 6							
1	0	0	10	10	0	0	0	0
13	0	0	10	10	0	0	0	0

c. Photographs



Image 1. Adult asparagus beetles in the crop.



Image 2. Asparagus beetle larvae in the crop.



Image 3. Experimental set-up showing Oasis® blocks with asparagus fern.

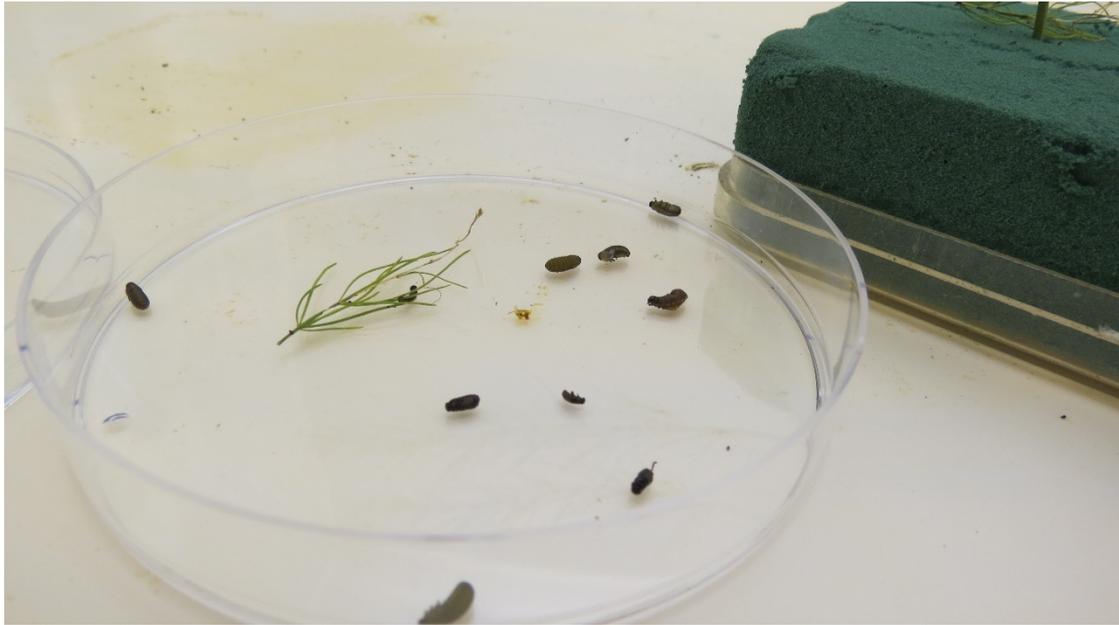


Image 4. Dead asparagus beetle larvae.



Certificate of

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

This certifies that

Warwick Crop Centre, School of Life Sciences

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

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

**Agriculture/Horticulture
Biologicals and Semiochemicals**

Date of issue: **6 October 2017**
Effective date: **20 March 2017**
Expiry date: **19 March 2022**

Signature

Aislin Richardson
Authorised signatory

Certification Number

ORETO 381



Chemicals Regulation Division



Department of
**Agriculture and
Rural Development**