





# Horticultural Fellowship Awards

Interim Report Form

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Project title:	Maintaining the expertise for developing and communicating practical Integrated Pest Management (IPM) solutions for Horticulture
Project number:	CP 89
Project leader:	Jude Bennison, ADAS
Report:	Interim, 30 April 2013
Previous report:	Interim report 2012
Fellowship staff:	Jude Bennison, Senior Entomologist, ADAS Boxworth (lead Fellowship mentor)
	Mike Lole, Senior Entomologist, ADAS Rosemaund (mentor)
	Steve Ellis, Senior Entomologist, ADAS High Mowthorpe (mentor)
	The late John Buxton, Senior Entomologist (mentor)
	John Atwood, Senior Horticultural Consultant (mentor)
	Chris Dyer, Statistician, ADAS (mentor)
	Heather Maher, Senior Research Manager, ADAS Boxworth (mentor until August 2012)
	Kerry Maulden, Senior Research Manager, ADAS Boxworth (mentor)
	Shaun Buck, Senior Research Manager, ADAS High Mowthorpe (mentor)
("Trainees")	Gemma Hough, Entomologist, ADAS

	Boxworth (Fellowship trainee Entomologist and Project Manager from Dec 2012)
	Tom Pope, Entomologist, ADAS Boxworth (Fellowship trainee Entomologist and Project Manager until August 2012)
	Gemma Gillies, Graduate Entomologist, ADAS Boxworth (Fellowship trainee Entomologist until Dec 2012)
	Tracie Evans, Research Technician, ADAS Boxworth (Fellowship trainee scientific support staff until August 2012)
	Chloe Whiteside, Research Technician, ADAS Boxworth (Fellowship trainee scientific support staff)
	Robert Drummond, Technician, ADAS Boxworth (Fellowship trainee scientific support staff)
	Abby Wood, Technician, ADAS Boxworth (Fellowship trainee scientific support staff)
Location of project:	ADAS Boxworth and commercial farms and nurseries
Industry Representative:	-
Date project commenced:	01 April 2011
Date project completed (or expected completion date):	31 March 2016

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# AUTHENTICATION

We declare that this work was done under our supervision according to the procedures described herein and that the report represents a true and accurate record of the results obtained.

Jude Bennison Senior Research Entomologist ADAS

Signature	Date
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Report authorised by:

Dr Tim O'Neill Horticulture Research Manager ADAS

Signature ..... Date .....

# CONTENTS

Progress Against Objectives1
Objectives1
Summary of Progress1
Milestones not being reached6
Do remaining milestones look realistic?6
Other achievements in the last year not originally in the objectives7
Changes to Project7
Are the current objectives still appropriate for the Fellowship?7
Grower Summary8
Headline8
Background8
Summary10
Financial Benefits13
Action Points13
Science Section15
Introduction15
Materials and methods17
Results and Discussion21
Conclusions
Knowledge and Technology Transfer33
Glossary
References

# **Progress Against Objectives**

# Objectives

Ob	ective	Original	Actual	Revised
		Completion	Completion	Completion
		Date	Date	Date
1.	Provide mentoring of two next generation ADAS research entomologists to equip them with the knowledge, skills, competencies and flexibility required to develop IPM strategies on horticultural crops.	31/03/2016	ongoing	-
2.	Deliver practical solutions to selected current and emerging pest management problems through specific applied research projects.	31/03/2016	ongoing	-
3.	Transfer knowledge and new IPM developments to the industry through a range of communication media.	31/03/2016	ongoing	-

# Summary of Progress

### Objective 1: Mentor two 'next generation' IPM research Entomologists

Tom Pope was already in post at ADAS Boxworth at the start of the Fellowship. He joined ADAS in 2009 and worked with Jude Bennison and colleagues on a range of projects investigating the biology and control of various horticultural pests including aphids, cabbage root fly and vine weevil. As part of the Fellowship Tom led work on predatory mites in soft fruit, biological control of vine weevil, incidence of aphid hyperparasitoids and biological control of aphids on outdoor lettuce. In August 2012, Tom left ADAS to join Harper Adams University as a lecturer in entomology and applied pest management research, where he is now training future entomologists. Tom is now a valued research collaborator with ADAS, already working with Jude Bennison and her team in two Defra-funded IPM projects.

Gemma Gillies joined ADAS Boxworth in October 2011 and assisted on the Fellowship projects, taking over the work on biological control of vine weevil in August 2012. Gemma left ADAS to return to teaching in December 2012 and ADAS has now recruited to replace her in its pest management team. The new ADAS Entomologist will start at ADAS Boxworth on 7 May 2013.

Gemma Hough joined ADAS Boxworth as a research entomologist in December 2012 after completing a HDC-funded PhD studentship on the biology and control of currant lettuce aphid at Warwick University. As part of the Fellowship during 2013 Gemma will be taking over work on biological control of vine weevil, biological control of aphids on lettuce and monitoring hyperparasitism in HNS. She is already involved in two HDC-funded projects, on improving biological control of aphids on protected herbs (PE 006a) and on reducing damage by *Scaptomyza flava* on baby-leaf salads (FV 408).

Mentoring activities during the second year of the Fellowship included:

#### Visits to commercial nurseries and farms

Visits were made by Gemma Gillies and Gemma Hough with Senior ADAS entomologists, Jude Bennison and the late John Buxton and with ADAS horticultural consultants, David Talbot and Angela Huckle. Nurseries and farms visited included:

Protected ornamentals: Gemma Gillies made consultancy visits to discuss IPM strategies with the late John Buxton.

Hardy nursery stock: Gemma Gillies made consultancy visits and specific monitoring of aphids and parasitoids with Jude Bennison and the late John Buxton. Gemma Hough visited growers with ADAS HNS consultant David Talbot to discuss leaf and bud nematode problems and potential control methods within IPM programmes.

Soft fruit: Gemma Gillies visited soft fruit nurseries with Jude Bennison to monitor for thrips, predatory mites, predatory bugs and aphid parasitoids and hyperparasitoids on protected strawberry.

Field vegetables: Gemma Gillies visited outdoor cucurbit growers with ADAS field vegetable consultant Angela Huckle to identify pests and take photographs for the HDC Crop Walkers Guide on outdoor cucurbits.

Protected herbs: Gemma Hough visited protected herbs growers with Jude Bennison to discuss aphid problems and biocontrol strategies.

#### Pest and biocontrol agent identification

Laboratory training on identification of key horticultural pests was completed by Gemma Gillies, Gemma Hough and Tom Pope as well as key members of the scientific support team at ADAS Boxworth. Training courses included:

- Aphid parasitoid and hyperparasitoid spp. identification (training given by Tom Pope and Tracie Evans)
- Scaptomyza spp. Identification (training given by Heather Maher)
- Thrips spp. identification (training given by Mike Lole and Jude Bennison)
- Predatory mite spp. identification (training given by Mike Lole)
- Free living nematode spp. identification (training given by Heather Maher and Shaun Buck)

#### Technical updates on biocontrol agents, biopesticides, pesticides and horticultural research

Technical meetings with suppliers of pesticides, biopesticides and biocontrol agents were attended throughout the year. These meetings provided updates on new products under development or those recently available for use by UK growers. Industry commodity group meetings and HDC research update meetings were also attended, where the trainees discussed key pest problems and research needs with growers. These included the BLSA (British Leafy Salads Association) conference, BHTA (British Herbs Trade Association) technical meeting and SPGA (Speciality Produce Growers Association) technical meeting. Scientific meetings attended included a Royal Entomological Society Meeting on 'Insects in a human-dominated world', the AAB (Association of Applied Biologists) conference on 'Advances in Biological Control'. Tom Pope and Gemma Gillies gave presentations at both these meetings.

Objective 2: Deliver practical solutions to selected current and emerging pest management problems through specific applied research projects

#### Efficacy of entomopathogenic nematodes against vine weevil

The efficacies of two commercially available nematode products for the control of vine weevil (VW) larvae in substrate-grown strawberry were compared: Nemasys® L (*Steinernema kraussei*) and the newly available patented product containing a mix of *Steinernema feltiae*, *Steinernema carpocapsae* and *Heterorhabditis megidis* (SuperNemos®).

The experiment was done in a poly tunnel at ADAS Boxworth. On 1 May 2012, ten barerooted everbearer strawberry plants were planted per standard one metre-long grow-bag (80% peat and 20% wood fibre). VW eggs were added on 1 August (15 eggs per plant) and curative applications of the nematode products were made on 6 September. In late October, plants were destructively sampled and the numbers of live larvae in each grow-bag were recorded.

The highest numbers of larvae were recorded in the untreated grow-bags which had a mean of 61.5 larvae per bag (equivalent to 6.2 larvae per plant). Plants treated with Nemasys L and SuperNemos both reduced the mean number of larvae per bag to 3.7 and 6.2 respectively (equivalent to 0.4 and 0.6 per plant respectively), which were both significantly different to the untreated control. Nemasys L and SuperNemos were equally effective, giving 94% and 90% control of VW larvae compared with untreated plants.

#### Aphid hyperparasitoids on protected edibles, soft fruit and ornamentals

Aphid hyperparasitoids were collected from a hardy nursery stock (HNS) site in Norfolk where the grower used regular releases of the newly available aphid parasitoid mix, which included the six parasitoid species *Aphidius colemani*, *Aphidius* ervi, Aphelinus *abdominalis*, *Aphidius matricariae*, *Praon volucre* and *Ephedrus cerasicola*.

The site was sampled on two occasions and parasitised (mummified) aphids were collected. Where possible, the aphid species and primary parasitoid genus were identified from the appearance of the 'mummy'. Evidence of primary parasitoid adult emergence (indicated by a neat circular exit hole) or hyperparasitoid adult emergence (indicated by a ragged emergence hole) was also recorded. Where there was no emergence hole, the mummified aphids were kept in the laboratory until either a primary or a hyperparasitoid adult emerged.

On the first sampling date percentage hyperparasitism was 33-50% and only one hyperparasitoid species was present (*Asaphes* sp). On the second sampling date, percentage hyperparasitism was17-70%. Three species of hyperparasitoids emerged from mummies collected on this sampling date: *Dendrocerus* sp. *Asaphes* sp. and *Alloystra brevis*. The potato aphid, *Macrosiphum euphorbiae*, the violet aphid, *Myzus ornatus* and the peach-potato aphid, *Myzus persicae* were the only parasitised aphids recorded.

### Biological control of aphids on lettuce

The population dynamics of aphids in response to the release of parasitoids in an organic lettuce crop were monitored. In addition, any hyperparasitism was recorded to determine whether it could affect the aphid control provided by primary parasitoids.

Fifty lettuce plants were assessed in a new planting on five weekly occasions during the summer. For each plant the number and species of aphids, the number of parasitised

'mummies' and any aphid predators or pathogens were recorded. Evidence of primary parasitoid emergence or hyperparasitoid emergence was also recorded. Where there was no emergence hole, the mummified aphids were kept in the laboratory until either a primary or a hyperparasitoid adult emerged. Parasitoids were released into the field containing our monitoring planting by the grower on 20, 27 June and 4 July 2012 at 0.23, 0.35 and 0.47 parasitoids/m<sup>2</sup> respectively. After the conclusion of the weekly monitoring, the farm was visited again on 21 August where two additional lettuce crops where no parasitoids had been released were monitored.

All the aphids recorded during the weekly monitoring were *Myzus persicae*. Aphid populations peaked on 3 July at 10 aphids per plant which was followed by a population crash. The decline in aphids coincided with a significant number of the aphids being infected with naturally-occurring entomopathogenic fungi. During the monitoring period in June and July only five mummies were found and of these, 80% were hyperparasitised, 50% of which were identified as *Asaphes* spp. On the additional monitoring date in August natural parasitism of both *M. persicae* and *M. euphorbiae* was observed. Fifteen mummies were recorded and 63% of these were hyperparasitised. The species responsible were identified as *Asaphes*, *Alloxysta* and *Dendrocerus* spp.

### Review of alternatives to Vydate for the control of leaf and bud nematodes

A review of the literature indicated that programmes of high crop hygiene remain the most effective cultural control measure against leaf and bud nematodes. Hot water treatments can also be effective but are not used as the requirements for and safety to all susceptible HNS species and cultivars are not available. Currently there are no effective alternative nematicides to oxamyl (Vydate 10G) for the control of this pest, but the review identified some potential alternative control measures that justify evaluation.

#### Objective 3: Transfer knowledge of new IPM developments to the industry

Knowledge transfer activities delivered by the trainees in year 2 of this project related both to this Fellowship project, and also to other horticultural projects, and included:

Publications (with input from experienced ADAS colleagues):

• HDC News articles on the Entomology Fellowship (CP 89) and the leaf miner *Scaptomyza flava* (FV 408), April 2013 (Gemma Hough)

• HDC Factsheet 10/12 Midge, mite and caterpillar pests of cane fruit crops (Tom Pope).

• Defra RADAR - Autumn 2012. Title: A new way of tackling an old problem (report on CRD-funded project evaluating the potential of using refuge traps as a means of disseminating entomopathogenic fungi for the control of adult vine weevil), Tom Pope.

• Bennison, J., Pope, T., Greetham, J., Evans, T. & Maher, H. (2012) Improved biological control of 'problem' aphids on protected herbs. IOBC/wprs Bulletin. 80:155-158

Industry Presentations:

• Summary of results on HDC-funded project FV 408: Baby-leaf Cruciferae and Watercress: Improved control of *Scaptomyza flava* at SPGA Technical Meeting (Gemma Hough and Jude Bennison)

• Summary of the Fellowship project CP 89 at ADAS Technical Skills Meeting (Gemma Hough)

• Summary of Entomology work at ADAS at Syngenta Horticultural meeting (Gemma Gillies)

Scientific Conference Presentations:

• Royal Entomological Society Meeting - Insects in a human dominated world- The Horticultural Fellowship explained - Summary of the Fellowship project CP 89 (Gemma Gillies).

• Royal Entomological Society Meeting - Insects in a human dominated world- Are adult vine weevils running out of places to hide? – results from CRD-funded project PS2134 (Tom Pope).

• AAB Advances in Biological Control meeting; Aphid parasitoids - new opportunities and challenges – results of HDC-funded projects on protected herbs, PE 006 and PE 006a (Tom Pope).

• AAB Advances in Biological Control meeting; The ADAS IPM Horticultural Fellowship (Gemma Gillies).

### Milestones not being reached

None

### Do remaining milestones look realistic?

Yes

# Other achievements in the last year not originally in the objectives

Trainees have worked with experienced ADAS entomologists on a wide range of horticultural projects over the last year. These included:

- HDC-funded project PE 006a Protected herbs: improved biological control of aphids.
- HDC-funded project FV407- Baby-leaf Cruciferae and Watercress: Improved control of Scaptomyza flava.
- HDC Crop Walkers Guide pests and diseases of outdoor cucurbits.
- Updating the Best-Practice Guide to Integrated pest and disease management on protected herbs on the HDC website.
- CRD-funded project PS2134 Use of refuge traps to disseminate entomopathogenic fungi for the control of adult vine weevil.
- HortLINK project HL001107 Biological, semiochemical and selective chemical management methods for insecticide resistant western flower thrips on protected strawberry.
- Defra-funded project TH0102 Improving control of oak processionary moth.
- Defra-funded project FFG 1146 Tree health: review and analysis of control strategies for established pests and pathogens of trees to inform current and future management.
- Defra-funded project Combating Resistance to Aphicides in UK Aphid Pests.
- CRD-funded report Pest, weed and disease incidence report 2012

In addition to the technical skills learnt through involvement on these projects, this work has provided several knowledge transfer opportunities as previously discussed. These activities were delivered by Tom Pope, Gemma Hough and Gemma Gillies.

# **Changes to Project**

### Are the current objectives still appropriate for the Fellowship?

Indicate any changes to the ordinal objectives that you would like to make and provide any information that you can to support this decision.

#### None

# **GROWER SUMMARY**

# Headline

- The entomopathogenic nematode products Nemasys L and SuperNemos were equally effective and significantly reduced numbers of live vine weevil larvae in substrate-grown strawberry when compared with untreated controls.
- Aphid hyperparasitoids were identified on protected HNS and on outdoor organic lettuce where growers were releasing aphid parasitoids during 2012.
- A literature review to identify potential alternatives to oxamyl (Vydate 10G) for leaf and bud nematode control on HNS indicated that a high standard of nursery hygiene remains the most effective cultural control measure but potential alternative nematicides and cultural methods were identified.

# Background

## Efficacy of entomopathogenic nematodes against vine weevil

Vine weevil (VW, *Otiorhynchus sulcatus*) remains one of the most serious problems in both soft fruit and nursery stock industries. In order to reduce damage caused by this pest, controls can be targeted against both the larvae in the soil and the adult weevils within the crop. Biological control of VW is preferable to the use of insecticides in Integrated Pest Management (IPM) programmes. Current options for biological control of VW larvae are entomopathogenic nematodes (various species and products) and the entomopathogenic fungus *Metarhizium anisopliae* (Met52).

This experiment compared the efficacies of two commercially available nematode products for the control of VW larvae in substrate-grown strawberry: Nemasys® L (*Steinernema kraussei*) and the newly available patented product containing a mix of *Steinernema feltiae*, *Steinernema carpocapsae* and *Heterorhabditis megidis* (SuperNemos®).

### Aphid hyperparasitoids on protected ornamentals

Aphid parasitoids are widely used for biological control of aphids within IPM programmes on many protected crops. Until recently, biological control of aphids on protected crops relied mainly on three aphid parasitoid species:

- Aphidius colemani for control of e.g. the peach-potato aphid, Myzus persicae and the melon-cotton aphid, Aphis gossypii.
- Aphidius ervi and Aphelinus abdominalis for control of e.g. the potato aphid, Macrosiphum euphorbiae and the glasshouse-potato aphid, Aulacorthum solani.

Use of aphid parasitoids on some crops has increased recently, due to the availability of a new mix of six parasitoid species. The new mix contains the above three parasitoid species plus an additional three species (*Aphidius matricariae*, *Ephedrus cerasicola* and *Praon volucre*) which has extended the range of aphid species that can be parasitised, and has thus led to further uptake of aphid parasitoids on a range of crops. In 2005, in a MAFF (now Defra)-funded project on developing IPM in outdoor Hardy Nursery Stock (HNS), ADAS confirmed that hyperparasitoids (secondary parasitoids which parasitise the primary aphid parasitoids) were a potential problem in naturally- parasitised aphids in outdoor HNS (Buxton *et al.* 2005). More recent investigations by Rob Jacobson in HDC-funded project PC 295, 295a and 295b have shown that breakdown in aphid control by parasitoids in mid-summer on some sweet pepper nurseries were predominantly due to the presence of hyperparasitoids (Jacobson 2010, 2011).

During 2011 in this Fellowship project, the presence of hyperparasitism was monitored and confirmed in sweet pepper, protected strawberry and HNS crops. A range of aphid species were parasitised by both *Aphidius* spp. and *Praon* spp. The hyperparasitoid species indentified were similar to those recorded in PC 295 and 295a and b, including *Asaphes suspensus*, *Asaphes vulgaris*, *Dendrocerus carpenteri*, *Dendrocerus laticeps* and *Pachyneuron* sp. On protected strawberry, HNS and sweet pepper hyperparasitism reached 5, 32 and 25% respectively. The aim during 2012 was to continue monitoring hyperparasitism at a HNS site.

#### Biological control of aphids on lettuce

Control of aphids on lettuce with pesticides is becoming increasingly difficult due to the limited number of pesticides available, pressures to reduce pesticide use and the increasing aphid resistance issues to both insecticides and to resistant cultivars which have been observed on lettuce for the peach-potato aphid, *Myzus persicae* and for currant-lettuce aphid, *Nasonovia ribisnigri* respectively. A major grower has reported achieving successful control of aphids in organic outdoor lettuce through the release of parasitoids. The use of biological control in field-grown lettuce, particularly for organic growers, could be an important component of an IPM programme.

Following discussion with the HDC and members of the British Leafy Salad Association, it was decided to evaluate the population dynamics of aphids in response to the release of parasitoids in an organic lettuce crop and to determine whether hyperparasitism occurred and if this could threaten the aphid control provided by primary parasitoids.

#### Review of the control of leaf and bud nematodes

Leaf and bud nematodes (LBN), *Aphelenchoides sp.*, are a significant foliar pest of the hardy nursery stock plants whose feeding results in angular-shaped dark blotches on the leaves which are delineated by the veins and often accompanied by leaf distortion. Subsequent damage from a LBN infestation can make a plant unmarketable causing significant economic losses for growers. Furthermore, once present on a nursery it is a challenging pest to eradicate due to its transmission being facilitated by overhead irrigation and its ability to survive for several years in infested dried leaf debris. Currently the only effective nematicide against this pest in the UK is oxamyl (Vydate 10G), which has an EAMU for use on protected HNS. Not all growers wish to use Vydate as it is not compatible with biological control agents used for other pests within IPM programmes and its use requires precautions for operator and environmental protection, a re-entry time to treated glasshouses and a harvest interval. Many growers prefer to use stringent nursery hygiene methods and sub-irrigation as key cultural control methods for the pest. This review aimed to summarise and collate potential alternative control measures available for LBN.

### Summary

#### Efficacy of entomopathogenic nematodes against vine weevil

The efficacies of two commercially available nematode products for the control of vine weevil (VW) larvae in substrate-grown strawberry were compared: Nemasys® L (*Steinernema kraussei*) and the newly available patented product containing a mix of *Steinernema feltiae*, *Steinernema carpocapsae* and *Heterorhabditis megidis* (SuperNemos®).

The experiment was done in a poly tunnel at ADAS Boxworth. On 1 May 2012, ten barerooted everbearer strawberry plants were planted per standard one metre-long grow-bag (80% peat and 20% wood fibre). VW eggs were added on 1 August (15 eggs per plant) and curative applications of the nematode products were made on 6 September. In late October, plants were destructively sampled and the numbers of live larvae in each grow-bag were recorded.

The highest numbers of larvae were recorded in the untreated grow-bags which had a mean of 61.5 larvae per bag (equivalent to 6.2 larvae per plant). Plants treated with Nemasys L and SuperNemos both reduced the mean number of larvae per bag to 3.7 and 6.2 respectively (equivalent of 0.4 and 0.6 per plant respectively), which were both significantly different to the untreated control. Nemasys L and SuperNemos were equally effective, giving 94% and 90% control respectively of VW larvae compared with untreated plants. There was no statistical difference between the control provided by Nemasys L and SuperNemos.

#### Aphid hyperparasitoids on protected ornamentals

Aphid hyperparasitoids were collected from a hardy nursery stock site in Norfolk where the grower used regular releases of a new aphid parasitoid mix, which included the six parasitoid species *Aphidius colemani*, *Aphidius ervi* and *Aphelinus abdominalis*, *Aphidius matricariae*, *Praon volucre* and *Ephedrus cerasicola*. The parasitoids were released weekly during the sampling period and the '6-pack' mix was supplemented with releases of single species as necessary e.g. with *Aphidius ervi* in 'hotspots' of the potato aphid, *Macrosiphum euphorbiae*.

The site was sampled on 18 May and 1 August and parasitised (mummified) aphids were collected. Where possible, the aphid species and primary parasitoid genus were identified from the appearance of the 'mummy'. Evidence of primary parasitoid adult emergence (indicated by a neat circular exit hole) or hyperparasitoid adult emergence (indicated by a ragged emergence hole) was also recorded. Where there was no emergence hole, the mummified aphids were kept in the laboratory until either a primary or a hyperparasitoid adult emerged.

On the first sampling date percentage hyperparasitism was 33-50% and only one hyperparasitoid species was present (*Asaphes* sp). On the second sampling date, percentage hyperparasitism was 17-70%. Three species of hyperparasitoids emerged from mummies collected on this sampling date: *Dendrocerus* sp. *Asaphes* sp. and *Alloystra brevis*. The potato aphid, *Macrosiphum euphorbiae*, the violet aphid, *Myzus ornatus* and the peach-potato aphid, *Myzus persicae* were the only parasitised aphids recorded.

#### Biological control of aphids on lettuce

Following discussions with a large lettuce grower who had been achieving successful control of aphids in organic outdoor lettuce through the release of parasitoids, it was decided to evaluate the population dynamics of aphids in response to the release of parasitoids in an organic lettuce crop and to determine whether hyperparasitism occurred and whether this could threaten the aphid control provided by primary parasitoids.

A new lettuce planting was monitored on five weekly occasions during the summer. Sampling started on 20 June, one week after the crop was planted. Fifty plants were assessed and for each plant the number and species of aphids, the number of mummies and any aphid predators or pathogens were recorded. Evidence of primary parasitoid emergence or hyperparasitoid emergence was also recorded. Where there was no emergence hole, the mummified aphids were kept in the laboratory until either a primary or a hyperparasitoid adult emerged. The grower released *Aphidius colemani* into the field containing the monitored planting on 20, 27 June and 4 July 2012 at 0.23, 0.35 and 0.47 parasitoids/m<sup>2</sup> respectively. After the conclusion of the weekly monitoring, the farm was visited again on 21 August where two additional lettuce crops where no parasitoids had been released were monitored.

All the aphids recorded during the weekly monitoring were *Myzus persicae*. Aphid populations peaked on 3 July at a mean of 10 aphids per plant which was followed by a population crash. The decline in aphids coincided with a significant number of the aphids being infected with naturally-occurring entomopathogenic fungi. During the monitoring period in June and July only five mummies were recorded, of which 80% were hyperparasitised, 50% of which were identified as *Asaphes* spp. On the additional monitoring date natural parasitism was observed of both *M. persicae* and *M. euphorbiae*. Fifteen mummies were recorded, of which 63% were hyperparasitised. The species responsible were identified as *Asaphes*, *Alloxysta* and *Dendrocerus* spp.

#### Review of the control of leaf and bud nematodes

Leaf and bud nematodes, *Aphelenchoides sp.*, are a significant foliar pest of hardy nursery stock plants whose feeding results in angular-shaped blotches on the leaves which are delineated by the veins and often accompanied by leaf distortion. Subsequent damage from a LBN infestation can make a plant unmarketable causing significant economic losses for growers. Furthermore, once present in a nursery it is a challenging pest to eradicate due to

its transmission being facilitated by overhead irrigation and its ability to survive for several years in infested dried leaf debris.

This review summarised and collated current and potential alternative control measures for LBN. The review showed that currently the only effective nematicide against this pest in the UK is oxamyl (Vydate 10G) and that stringent nursery hygiene methods and sub-irrigation are key cultural control methods. The potential use and further evaluation of hot water treatments, biological control (bacteria and entomopathogenic nematodes), natural plant extracts/biopesticides and host plant resistance were also discussed.

# **Financial Benefits**

- Biocontrol of aphids usually requires regular releases of parasitoids. High proportions of aphid hyperparasitoids reduce the effectiveness of these parasitoids, resulting in increased losses caused by aphids. Growers will benefit from being aware of this risk on a range of horticultural crops so that they can adapt their IPM programmes if needed.
- Growers are not always confident of using entomopathogenic nematodes for control of vine weevil in strawberry, and are unsure of which product to buy. Growers will benefit from the results in this project that demonstrated that a new nematode product, SuperNemos was equally effective in controlling vine weevil in substrate-grown strawberry as one of the 'standard' products, Nemasys L. Further work will be done in this project during 2013 to compare control of vine weevil by all nematode products available in the UK and by the entomopathogenic fungus Met52.
- Not all growers wish to use oxamyl (Vydate 10G) for control of leaf and bud nematode on HNS, preferring to use high standards of nursery hygiene together with sub-irrigation as key cultural control methods. Growers will benefit from the literature review in this project which identified some potential alternative methods for control which justify consideration for future research.

# **Action Points**

 When selecting nematode products for control of vine weevil, growers should consider their optimum temperature range and cost in addition to available information on comparative efficacy.

- Growers using aphid parasitoids in any crop should be aware that aphid hyperparasitism may occur. Look out for ragged emergence holes in aphid 'mummies' as an indicator that hyperparasitoids are present and monitor percentage parasitism and hyperparasitism.
- Seek advice from your biocontrol supplier or IPM consultant if percentage aphid hyperparasitism starts to increase. You may need to switch from using aphid parasitoids to aphid predators, and/or IPM-compatible pesticides.
- Vydate 10G is the only effective nematicide currently available for control of leaf and bud nematodes in HNS. This pesticide is not compatible with biological control agents used within IPM. Maintaining high standards of crop hygiene together with using subirrigation is currently the most effective cultural control measure for this pest.

# SCIENCE SECTION

# Introduction

#### Efficacy of entomopathogenic nematodes against vine weevil

Vine weevil (VW, *Otiorhynchus sulcatus*) remains one of the most serious problems in both soft fruit and nursery stock industries. In order to reduce damage caused by this pest, controls can be targeted against both the larvae in the soil and the adult weevils within the crop. Biological control of VW is preferable to the use of insecticides in Integrated Pest Management (IPM) programmes. Current options for biological control of VW larvae are entomopathogenic nematodes (various species and products) and the entomopathogenic fungus *Metarhizium anisopliae* (Met52).

The aim of this project was to compare the efficacies of two commercially available nematode products for the control of VW larvae in substrate-grown strawberry: Nemasys® L (*Steinernema kraussei*) and the newly available patented product containing a mix of *Steinernema feltiae*, *Steinernema carpocapsae* and *Heterorhabditis megidis* (SuperNemos®).

### Aphid hyperparasitoids on protected edibles, soft fruit and ornamentals

Aphid parasitoids are widely used for biological control of aphids within IPM programmes on many protected crops. Until recently, biological control of aphids on protected crops relied mainly on three aphid parasitoid species:

- Aphidius colemani for control of e.g. the peach-potato aphid, *Myzus persicae* and the melon-cotton aphid, *Aphis gossypii*.
- Aphidius ervi and Aphelinus abdominalis for control of e.g. the potato aphid, Macrosiphum euphorbiae and the glasshouse-potato aphid, Aulacorthum solani.

Use of aphid parasitoids on some crops has increased recently, due to the availability of a new mix of six parasitoid species. The new mix contains the above three parasitoid species plus an additional three species (*Aphidius matricariae*, *Ephedrus cerasicola* and *Praon volucre*). The mix is produced by Viridaxis in Belgium and is available as various products, such as Aphidsure mix ® (for use on various crops) and Aphidsure fragaria ® (for strawberry) supplied by BCP Certis and FresaProtect ® (for strawberry) and OrnaProtect ® (for ornamentals) from various other suppliers. These products have extended the range of

aphid species that can be parasitised, and have thus led to further uptake of aphid parasitoids on a range of crops, particularly those such as HNS and soft fruit that can be attacked by a wide range of aphid species.

In 2005, in a MAFF (now Defra)-funded project on developing IPM in outdoor HNS, ADAS confirmed that hyperparasitoids (secondary parasitoids which parasitise the primary aphid parasitoids) were a potential problem in naturally- parasitised aphids in outdoor HNS (Buxton *et al.* 2005). Seven species of hyperparasitoids were confirmed in this project. More recent investigations by Rob Jacobson in HDC-funded project PC 295, 295a and 295b have shown that breakdown in aphid control by parasitoids in mid-summer on some sweet pepper nurseries were predominantly due to the presence of hyperparasitoids (Jacobson 2010, 2011).

During 2011 in this Fellowship project the presence of hyperparasitism was monitored and confirmed in sweet pepper, protected strawberry and hardy nursery stock crops. A range of aphid species were parasitised by both *Aphidius* spp. and *Praon* spp. The hyperparasitoid species indentified were similar to those recorded in PC 295 and 295a and b, including *Asaphes suspensus*, *Asaphes vulgaris*, *Dendrocerus carpenteri*, *Dendrocerus laticeps* and *Pachyneuron* sp. On protected strawberry, hard nursery stock (HNS) and sweet pepper hyperparasitism reached 5, 32 and 25%.

The aim during 2012 was to continue monitoring hyperparasitism at a HNS site

### Biological control of aphids on lettuce

Control of aphids on lettuce with pesticides is becoming increasingly difficult due to the limited number of pesticides available, pressures to reduce pesticide use and the increasing aphid resistance issues to both insecticides and to resistant cultivars that have been observed on lettuce for the peach-potato aphid, *Myzus persicae* and to currant-lettuce aphid, *Nasonovia ribisnigri* respectively. A major grower (G's) has reported achieving successful control of aphids in organic outdoor lettuce through the release of parasitoids. The use of biological control in field-grown lettuce, particularly for organic growers, could be an important component of an Integrated Pest Management (IPM) programme.

Following discussion with the HDC and members of the British Leafy Salad Association, it was decided to evaluate the population dynamics of aphids in response to the release of parasitoids in an outdoor organic lettuce crop and to determine whether hyperparasitism was present and if it could threaten the aphid control provided by primary parasitoids.

#### Review of the control of leaf and bud nematodes

Leaf and bud nematodes (LBN), *Aphelenchoides sp.*, are a significant foliar pest of hardy nursery stock plants whose feeding results in angular-shaped blotches on the leaves which are delineated by the veins and often accompanied by leaf distortion. Subsequent damage from a LBN infestation can make a plant unmarketable causing significant economic losses for growers. Furthermore, once present in a nursery it is a challenging pest to eradicate due to its transmission being facilitated by overhead irrigation and its ability to survive for several years in infested dried leaf debris. Currently the only effective nematicide against this pest in the UK is oxamyl (Vydate 10G), which has an EAMU for use on protected HNS. Not all growers wish to use Vydate as it is not compatible with biological control agents used for other pests within IPM programmes and its use requires precautions for operator and environmental protection, a re-entry time to treated glasshouses and a harvest interval. Many growers prefer to use stringent nursery hygiene methods and sub-irrigation as key cultural control methods for the pest. This review aimed to summarise and collate potential alternative control measures available for LBN.

# Materials and methods

#### Efficacy of entomopathogenic nematodes against vine weevil

The experiment consisted of three treatments (Table 1).

Table 1	Treatments,	rates and	methods	of application
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Trt. no	Product name	Active ingredient	Supplier	Recomm- ended rate	Equiv. nematode rate per litre compost	Application method
1	Untreated	-	-	-	-	-
2	Nemasys L	Steinernema kraussei	Becker Underwood	25,000 per plant	10,000 per I	Drench
3	SuperNemos	Steinernema carpocapsae +Steinernema feltiae + Heterorhabditis megidis	Flowering Plants Ltd	500,000 per m <sup>2*</sup>	5,000 per I	Drench

\*equivalent to 12,500 nematodes per plant when there are 10 plants per grow-bag (as in this experiment)

Experimental plants and substrate: Standard one metre-long grow-bags, each containing 25 litres of substrate (80% peat and 20% wood fibre), were obtained from Bulrush Horticulture Ltd and bare-rooted everbearer strawberry plants (cv. Calypso) were purchased from R. W. Walpole Ltd.

Experiment design: Ten strawberry plants were planted per grow-bag on 1 May. Each growbag represented a treatment plot, and each treatment had four replicates which were arranged in a randomised block design in a poly tunnel at ADAS Boxworth, Cambridgeshire (Figure 2).

Irrigation and temperatures: Overhead irrigation was used to establish the plants; those which did not establish were replaced on 8 May. Automatic drip irrigation was used thereafter. Temperature of the substrate at root depth was measured throughout the trial using two identical data loggers.



Figure 2 Strawberry experiment in grow-bags in a poly tunnel at ADAS Boxworth

Vine weevil egg infestation: On 1 August, 15 VW eggs were washed onto the soil around the stem of each plant.

Nematode applications: On 6 September, curative applications of each nematode product were applied as per supplier's recommendations (Table 1 and leaflet supplied with nematodes). Nematodes were applied with a syringe rather than a sprayer, to ensure dose accuracy to each plant.

Assessment of vine weevil larvae and plant vigour: On 23 and 24 October, plants were destructively sampled and the numbers of live VW larvae were recorded in each grow-bag by carefully searching through the roots and substrate. Visual assessments were also made of plant vigour (plant size and foliage health) before destructive sampling, using a scale of 0-5 where five was healthy and zero was dead.

Control of other pests and diseases: Regular applications of fungicides and biological control agents were applied as per commercial practice to control diseases, e.g. crown rot, and other pests, e.g. aphids, spider mites and thrips. The biological control agents used included the predatory mite *Neoseiulus (Amblyseius) cucumeris* for thrips control, a mix of six aphid parasitoid species for aphid control and the predatory mite *Phytoseiulus persimilis* for spider mite control. Fungicides applied are listed in Table 3.

Table 3 Fungicides applied during the experiment					
Date	Active ingredient	Trade name			
1 June 2012	Dimethomorph	Paraat			
11 June 2012	Myclobutanil	Systhane 20 EW			
	Boscalid + Pyraclostrobin	Signum			
26 June 2012 + 6 August 2012	Potassium hydrogen carbonate + Organosilicone surfactant	Potassium hydrogen carbonate + Silwett 77			

Statistical analysis: Data on the numbers of live larvae and plant vigour for each treatment were subjected to analysis of variance (ANOVA)

### Aphid hyperparasitoids on protected hardy nursery stock

Site selection: Parasitised aphids were collected from a HNS site in Norfolk (Darby Nursery Stock) where the grower used regular releases of the aphid parasitoid mix of six species (Table 4). The parasitoids were released weekly during the sampling period and the '6-pack' mix was supplemented with releases of single species as necessary e.g. with *Aphidius ervi* in 'hotspots' of the potato aphid, *Macrosiphum euphorbiae*.

 Table 4. Primary parasitoid species released.

Parasitoid species Aphidius ervi Aphidius colemani Aphidius matricariae Praon volucre Ephedrus cerasicola Aphelinus abdominalis

Sampling and identification: aphids were sampled on two dates, 18 May and 1 August. Parasitised aphids on a range of host crops were recorded and collected in order to identify the species of aphid that had been parasitised and whether the parasitised mummy had been hyperparasitised. If an emergence hole was present in the mummy, a record was made whether it was characteristic of a primary parasitoid or hyperparasitoid (i.e. round or jagged respectively). Where no emergence hole was found, the mummies were placed in glass Petri dishes in the laboratory at approximately 20°C until either a primary parasitoid or

a hyperparasitoid adult emerged. Emerging hyperparasitoids were identified to species at the Natural History Museum. In addition, wherever possible aphid species and primary parasitoid genus (based on aphid mummy colour and morphology i.e. *Aphidius* sp., *Praon* sp. or *Aphelinus* sp. / *Ephedrus* sp.) was recorded.

#### Biological control of aphids on lettuce

The aim of this study was to record the numbers and species of aphids and the levels of parasitism on an outdoor organic lettuce crop where the grower was releasing *Aphidius colemani* for the control of aphids. The work was carried out on an organic lettuce crop at G's in Cambridgeshire.

Assessments: On 20 June, a new planting was selected and its position marked out in the field. Fifty plants were then assessed by walking from the edge of the planting to the centre in a 'W' pattern. Plants were sampled at random and for each plant the number and species of aphids, the number of mummies (noting whether they were *Aphidius*, *Praon* or *Ephedrus*/*Aphelinus* species by observing the mummy colour as pale brown, beige or black respectively) and any aphid predators or pathogens were recorded.

Where possible, up to ten mummies were collected from each plant in order to record the species of aphid that had been parasitised and whether the emergence hole was characteristic of a primary parasitoid or hyperparasitoid (i.e. round or jagged respectively). If no emergence hole was present, the mummies were placed in a petri dish and brought back to ADAS Boxworth where the adult parasitoids were allowed to emerge and the species identified. The planting assessments were made on 20, 27 June, 3, 10, 17 and 24 July (Figure 5).

*Parasitoid release:* The grower released *Aphidius colemani* weekly for the control of aphids. Parasitoids were released into the field containing our monitoring planting on 20, 27 June and 4 July 2012 at 0.23, 0.35 and 0.47 parasitoids/m<sup>2</sup> respectively. Once aphids were observed in the crop, the grower increased the preventive rate of 0.23 per m<sup>2</sup> by approximately 0.1 parasitoids /m<sup>2</sup> (1000/ha) in response to light aphid infestations while an additional 0.3 parasitoids/m<sup>2</sup> (3000/ha) were used for heavy infestations.



Figure 5 Monitored planting on 20 June (left) and 24 July 2012 (right) assessment dates.

Additional assessment: After the conclusion of the weekly monitoring, the farm was visited again on 21 August where two additional lettuce crops (one close to harvest and the other one week old) were assessed on a single date.

#### Controlling leaf and bud nematode

A review of the literature was carried out to summarise the current and potential alternative control measures for leaf and bud nematode (LBN). General internet searches and searches via Scopus of scientific literature relating to control measures of foliar and root nematodes (without any date restrictions) were carried out.

# **Results and Discussion**

#### Efficacy of entomopathogenic nematodes against vine weevil

Effects of treatment on the number of larvae: Analysis of the mean number of live VW larvae per grow-bag showed a highly significant effect of treatment. The highest numbers of larvae were recorded in the untreated grow-bags which had a mean of 61.5 larvae per bag (equivalent to a mean of 6.2 larvae per plant, Figure 6).

Nemasys L and SuperNemos both reduced the mean number of larvae per bag to 3.7 and 6.2 respectively (equivalent to means of 0.4 and 0.6 per plant respectively), which were both significantly different to the untreated control (P<0.05). Nemasys L and SuperNemos were statistically equally effective.



**Figure 6** Mean numbers of live vine weevil larvae per grow-bag with standard error (SE). The least significant difference (LSD) was used to determine any significant differences. Different letters above bars indicate a significant difference.

Effects of treatment on plant vigour: Analysis of plant vigour scores per plot showed that there was no effect of any treatment on vigour observed during the experiment (Figure 7). The untreated control scored 4.9 (5 was very healthy and 0 was dead) suggesting that more than a mean of 60 live larvae per grow-bag (equivalent to six larvae per plant) are required before immediate visible crop damage occurs. Further damage may have been observed if the plants had been assessed later when VW larvae had the opportunity to continue feeding until pupation in late spring the following year.



Substrate temperatures: The critical period for substrate temperatures for nematode activity was between the date of nematode application (6 September) and the date assessments were done on surviving vine weevil larvae (23 October). During this period, temperatures remained within the activity range of Nemasys L (5-30°C), but mean and minimum temperatures dropped below 10°C (lower limit for SuperNemos) on several dates during September and October (Figure 8 and Table 9). However, substrate temperature did not appear to affect the levels of control provided by the two nematode products.



**Figure 8** Mean average, maximum and minimum substrate temperatures recorded by two data loggers at root level throughout the experimental period

 Table 9
 The optimum temperature range for the nematode products used as per the manufacturer's instructions supplied with the products.

Product	Optimum temperature range (°C)
Nemasys L	5-30
SuperNemos	>10

Future work: A similar experiment will be done during 2013, including additional available nematode products (those containing *Heterorhabditis bacteriophora*) and the substrate-incorporated entomopathogenic fungus *Metarhizium anisopliae* product Met52.

#### Aphid hyperparasitoids on protected edibles, soft fruit and ornamentals

Mummified aphids were collected from a range of hardy nursery stock crops including *Abelia*, *Cistus*, *Coprosma*, *Coronilla*, *Dianthus*, *Escallonia*, *Hebe*, *Lonicera*, *Photinia*, *Pittosporum*, *Sollya* and *Tracheospermum* spp.

On 18 May the parasitised aphids were almost all the potato aphid, *Macrosiphum euphorbiae* except for peach-potato aphid, *Myzus persicae* and violet aphid, *Myzus ornatus* found on Cistus *x pulverulentus* 'Sunset' (Table 10). The aphids were parasitised by either *Aphidius* or *Praon* spp. Hyperparasitism ranged between 33 and 50% and only one hyperparasitoid species was present (*Asaphes* sp).

On 1 August, the highest numbers of mummies were collected from *Coronilla valentina* subsp. glauca 'Citrina' (Table 11). All of the parasitised aphids on all host crops sampled were *M. euphorbiae*, and these had been parasitised by either *Praon* sp. or *Aphidius ervi*. Three species of hyperparasitoids were recorded emerging from mummies collected on this sampling date: *Dendrocerus* sp. *Asaphes* sp. and *Alloystra brevis*. Percentage parasitism ranged between 17 and 70%. The most hyperparasitism occurred on *Hebe variegata* and the primary parasitoid species was identified as *A. ervi*. Parasitoid emergence from the mummies was >83%. The data confirms that hyperparasitism was widespread in a range of HNS crops during 2012.

HINS	crops	samp	lea	on ra	5 1	lay	201
Plant	No. mummified aphids collected	Aphid species	Primary parasitoid	Hyperparasitoid	% parasitoid emergence	% hyper parasitism	
Outdoor Eschallonia	3	M. euphorbiae	?	Asaphes sp.	100	33	
<i>Eschallonia</i> 'Peach Blossom'	1	M. euphorbiae	Aphidius sp.		100	0	
<i>Eschallonia</i> 'Peach Blossom'	4	M. euphorbiae	Aphidius ervi	Asaphes sp.	100	50	
Cistus x pulverulentus 'Sunset'	8	Myzus persicae and Myzus ornatus	Aphidius sp.		100	0	
Abelia sp.	2	M. euphorbiae	Praon sp		100	0	
Abelia sp.	3	M. euphorbiae	Aphidius ervi		100	0	

 Table 10 Numbers of parasitised aphids and percentage hyperparasitism on a range of

 HNS
 crops
 sampled
 on
 18
 May
 2012

**Table 11** Numbers of parasitised aphids and percentage hyperparasitism on a range ofHNS crops sampled on 1 August 2012.

Plant	No. mummified aphids collected	Aphid species	Primary parasitoid	Hyperparasitoid	% parasitoid emergence	% hyper parasitism
Coronilla sp.	5	M. euphorbiae	Praon sp.		100	0
Coronilla valentina subsp. glauca 'Citrina'	52	M. euphorbiae	Praon sp.	Dendrocerus sp. and Asaphes sp.	88	29
Hebe sp.	7	M. euphorbiae			100	29
Hebe variegata	10	M. euphorbiae	Aphidius ervi	Asaphes sp., Dendrocerus and Alloysta brevis	100	70
Hebe variegata	13	M. euphorbiae	Aphidius ervi	Asaphes sp., Dendrocerus sp. and Alloxysta brevis	100	54
Hebe Margaret	10	M. euphorbiae	Aphidius ervi	Alloysta brevis	83	30
Coprosma 'Pacific night'	6	M. euphorbiae	Praon sp. and Aphidius ervi	Alloysta brevis	100	17

# Biological control of aphids on lettuce

Figure 12 shows the numbers of aphids which were observed at each monitoring date. All of the aphids recorded on the lettuce were *Myzus persicae* and their mean numbers increased to a peak of 10.2 per plant on 3 July. However, following this peak, aphid numbers declined until the last sampling date on 24 July. Rapid declines such as this are characteristic of aphid populations and are referred to in the literature as the 'mid-summer crash'. These population crashes commonly occur during mid-July, with a rapid decline in aphid numbers which continue to remain low for up to eight weeks (Karley *et al.*, 2004). Observed in both cropping and non cropping environments, various factors have been proposed to explain its occurrence which includes weather conditions, host-plant quality, natural enemies and mass emigration (Karley *et al.*, 2003).

During the monitoring period in this study, the decline in aphids coincided with a significant number of the aphids being infected with naturally-occurring entomopathogenic fungi, whose occurrence and spread during 2012 was likely to be promoted by the wet and humid conditions. Therefore, the fungal infection is considered to have made a significant contribution to the observed decline in aphid numbers. Aphid populations crashes have been observed in other studies in response to a fungal epizootic (Nielsen and Hajek, 2005).

During the monitoring period in June and July only five mummies were found and a total 80% hyperparasitism was observed, 50% of which were identified as *Asaphes* spp. The first parasitised aphid was recorded on 3 July followed by four additional mummies on 24 July. The one mummy collected on 3 July was hyperparasitised as were three of the four mummies collected on 24 July.

Although the host grower has reported successful use of aphid parasitoids on outdoor organic lettuce only low levels of parasitism were observed in this study. Low percentage parasitism was likely to have been due to the presence of the entomopathogenic fungi which killed most of the aphids that had infested the plants after planting.



**Figure 12** Number of winged and wingless aphids present at each monitoring date. *Aphidius colemani* was released on 20 June, 27 July and 4 July.

Following the weekly monitoring during June and July, an additional assessment was made in two lettuce crops on the same farm on 21 August. The grower had not yet made any parasitoid releases to these crops. From these crops 15 mummies were collected of which nine were healthy primary parasitoids and five were hyperparasitised as summarised in Table 13. The aphid species parasitised included *M. persicae* and the potato aphid, *M. euphorbiae.* These must have been naturally-occurring parasitoids, as the grower had only released *Aphidius colemani* to other crops on the farm, and this species does not parasitise *M. euphorbiae.* The *M. euphorbiae* mummies were confirmed as being parasitised by either *Aphidius ervi* or *Praon volucre* (Table 13). Parasitised *M. persicae* were hyperparasitised by *Asaphes* spp. and parasitised *M. euphorbiae* were hyperparasitised by *Asaphes*, *Alloxysta* and *Dendrocerus* spp. (Table 13). The species will be confirmed by the Natural History Museum and the results will be reported in the next annual Fellowship report.

**Table 13** Summary of the parasitoids and hyperparasitoids which emerged from mummiescollected on the additional assessment day (21 August).

Parasitised	Primary	Hyper	Counts
aphid	parasitoid	parasitoid	
M. euphorbiae	Praon volucre		1
M. persicae	Praon volucre		1
M. euphorbiae		Dendrocerus	1
		spp.	
M. persicae		Asaphes spp.	1
M. eurphorbiae		Alloxysta spp.	2
M. eurphorbiae		Asaphes spp	1
M. persicae	Aphidius spp.		2
M. euphorbiae	Aphidius ervi		1
M. persicae	Aphidius		1
	colemani		
M. euphorbiae	Aphidius spp.		1
M. persicae	Aphidius ervi		2

This study confirmed that hyperparasitism of aphid parasitoids occurred in the lettuce crop monitored and these have also been observed by the grower in previous years. Despite this, no reports of control failures due to hyperparasitism have yet been made.

Work will continue in 2013, to establish the effectiveness of aphid parasitoids in organic lettuce. Further crops where parasitoids are released will be monitored. Aphid numbers and species from planting to harvest and levels of parasitism and hyperparasitism will be recorded. Release of single or mixed parasitoid species will be compared and the role of other naturally-occurring aphid biological control agents will be quantified.

### Review of the control of leaf and bud nematodes

Leaf and bud nematodes (LBN), *Aphelenchoides* spp. are a significant foliar pest of the hardy nursery stock plants. These nematodes enter the leaves through the stomata and also infest the buds. Movement between plants and leaves is facilitated by the presence of a film of water, provided by either rainfall or overhead irrigation.

In the UK, *Aphelenchoides ritzemabosi* and *Aphelenchoides fragariae* are the two main nematode species of economic importance whose feeding results in angular-shaped blotches on the leaves which are delineated by the veins and often accompanied by leaf distortion (Southey, 1978). As the symptoms of LBN infestation are similar to infection by

various pathogens, laboratory tests should be performed to confirm their presence. It is also important to remember that infested plants do not always exhibit symptoms.

Cultural control methods are an important component of the management of leaf and bud nematodes within IPM programmes. The most effective of these is a programme of high crop hygiene as LBN can survive for several years in infested dried leaf debris. Cultural control programmes should include the removal and destruction of infested plants and debris, sterilisation of pots and equipment, not replanting in contaminated land and avoiding the use of overhead irrigation and misting systems which create ideal conditions for nematode transmission (Young, 1997). New plants entering the nursery should also be quarantined until it is confirmed that they are free from nematodes.

In addition to this, hot water treatment can be used to reduce or eliminate infestations to provide clean mother plants for micropropagation (Young, 1997). Jagdale and Grewal (2004) proposed that a 90°C water soil drench in the autumn or early spring could reduce the infestations of foliar nematodes. Various studies have provided specific recommendations for hot water treatments of different plants including immersing chrysanthemums for 20 minutes at 110°F to control A. ritzemabosi (Kearns & Walton, 1934) and immersing bulbs of *Polianthes tuberosa* for 30 minutes at 57°C to reduce damage by Aphelenchoides besseyi (Cuc et al., 2010). Various recommendations also exist for controlling A. besseyi and A. fragariae in strawberry plants (OEPP/EPPO, 2012; Qiu et al., 1993). However, while hot water treatments have been proven to be effective, the required temperature and duration of the treatment varies between plant species and cultivars and significant resources would be needed to determine the requirements for each, in order to give effective control and to avoid phytotoxicity. A review of hot water treatments for various plants is available (Gratwick and Southey, 1986).

Other methods of control which are available include chemical control using nematicides and disinfectants, biological control, natural plant extracts/biopesticides and host plant resistance.

#### Chemical control

Until its withdrawal in the UK in 2007, Temik (aldicarb) was the most effective chemical used for the control of LBN. Dynamec (abamectin) was also found to be effective in the HDC project HNS 86 against *A. ritzemabosi* but was not as persistent or as reliable as Temik (Young, 2000). A similar study on *A. fragariae* found that abamectin and diazinon was effective on *Lamium* and *Phlox* but not *Azalea* or *Begonia* spp. (LaMondia, 1999). In the UK, diazinon has been withdrawn but abamectin is still approved on certain crops.

In response to the withdrawal of Temik, the HDC project HNS 131 evaluated a range of alternatives for the control and management of LBN. In this study, Dynamec was found to be ineffective in controlling LBN and it was proposed that Vydate 10G (oxamyl) was the most effective replacement product, although it was not as effective as Temik (Bennison, 2007). Currently, Vydate 10G can be used at a growers own risk on outdoor and protected ornamentals for the control of non indigenous leaf miner species as given on EAMU 2322/2012.

More recently, other potential nematicides have been identified including spirotetramat (Movento) which has been observed to be effective against the root feeding lesion nematode (*Pratylenchus vulnus*) in walnut orchards (DeBuse, 2011) and against the cereal cyst nematode (*Heterodera avenae*) in wheat (Smiley, 2011). Currently its potential effect on LBN is unknown but as it has systemic activity and has an EAMU for use on protected ornamentals for control of other pests, it justifies evaluation.

Research is also underway in the US to determine whether Agri-50, an organic pesticide (alginate polysaccharide) available in the UK (which does not need to be registered as a pesticide as it acts by mechanical action), can be used as a nematicidal soil drench. Agri-50 proved ineffective against LBN in HDC project HNS 131 (Bennison 2007).

In the USA, the fatty acids product Insecticidal Soap gave 72% control of leaf nematodes 48 days after treatment when used as a foliar spray on *Hosta spp.*, although it gave no control in previous *in vitro* laboratory tests when tested as a water suspension (Jagdale & Grewal, 2002). In the UK, the fatty acid product Savona is available and approved as a foliar spray for control of various pests including aphids, whiteflies and spider mites on both outdoor and protected ornamentals. Savona proved to be ineffective against LBN in HDC project HNS 131 (Bennison 2007).

Jet5 (peroxyacetic acid) is widely used as a disinfectant in UK protected horticultural crops, to clean floors and benches etc between crops for control of disease pathogens. The same disinfectant (trade name ZeroTol) has been shown in the USA to have good activity against the leaf nematode *A. fragariae* (Jagdale & Grewal, 2002). This work showed that the disinfectant killed 100% of the nematodes within 48 hours in water suspension laboratory tests, and when used as a foliar spray on *Hosta* spp., it gave 73% reduction in nematode numbers 48 days after treatment, with no evidence of phytotoxicity. Jet 5 was also tested by ADAS against the stem nematode, *Ditylenchus dipsaci* for the bulb industry in HDC Project BOF 49 (Lole, 2001). This *in vitro* laboratory work showed that 75% of the nematodes were killed within one hour. Iodophor/acid disinfectants (*FAM 30 / Antec Virudine*) were also tested against stem nematode in Project BOF 49. They were the most

effective treatment in this study, killing 100% of the nematodes within five minutes. Both these disinfectants could have potential for control of LBN in HNS.

### **Biological control**

In addition to various chemicals, HDC project HNS 131 also investigated the effectiveness of the entomopathogenic nematode *Steinernema carpocapsae* in controlling LBN when applied as a 5-spray programme at a rate of 500 million per 1000m<sup>2</sup> which proved to be ineffective (Bennison 2007). However, a more recent study in America which mixed 100 *S. carpocapsae* infected cadavers of the wax moth (*Galleria mellonella*) (25 g wet weight) into the potting medium as both a preventative and curative application suppressed *A. fragariae* after 30 and 45 days following treatment (Jagdale. & Grewal, 2008). In another study, both dead and live *S. carpocapsae* reduced a range of soil parasitic nematodes (*Criconemella, Hoplolaimus, Longidorus, and Rotylenchus*) when applied around boxwood plants in combination with the use of nematicide granules (Jagdale *et al.*, 2002).

In addition to research on using entomopathogenic nematodes (EPNs) for controlling pest nematode species, research has also been done on using the symbiotic bacteria present in EPNs currently used against other pests e.g. vine weevil and sciarid flies. The bacteria, e.g. *Xenorhabdus* spp. is released inside the insect gut once the nematodes have entered the insect host's body. It is the bacteria that kills the host, rather than the nematodes themselves. The bacteria itself has been shown to act as a biological nematicide if released in water or soil containing nematodes. Research has been done in the UK on the use of bacteria from EPNs for the control of various pests including root-knot nematodes (Tabil *et al*, 2003). Recent work at Ohio State University, by the same research group who evaluated fatty acids and disinfectants against foliar nematodes (Grewal and Jagdale), has shown that bacteria from the same EPNs is highly toxic to leaf and bud nematodes. Although EPNs are available in the UK, use of the symbiotic bacteria alone is not currently approved for use as a biopesticide.

Other potential biological control agents include *Bacillus subtilis* whose purL gene has demonstrated nematicidal activity *in vitro* against various nematode species including the LBN *Aphelenchoides. besseyi* (*Xia et al.*, 2011). However, whether *B. subtilis* would penetrate the leaf tissue to kill LBN in propagated plants, and whether the commercially available strain of *B. subtilis* available as the product Serenade (strain QST 713) would have activity against LBN is currently unknown.

#### Natural plant extracts/biopesticides

In HDC project HNS 131 two plant extracts were tested for potential control of LBN (Bennison 2007). Nemagold (a liquid extract of marigold, *Tagetes erecta*, seaweed and 'organic matter'), marketed in Spain and certain other countries as a biostimulant, but also for repelling and controlling soil-dwelling nematodes including cyst nematodes, root-knot nematodes and free-living nematodes. The product is not approved in the UK and it proved to be ineffective in controlling LBN when used as a foliar spray in a 3-spray programme. The second plant extract tested in HNS 131 was garlic which was applied as repeated foliar applications but proved ineffective in controlling LBN. Other potential alternative methods for the control of LBN identified in HNS 131 were:

• Compost teas: There is evidence from use of compost teas (contain various active ingredients including bacteria and fungi) in the Netherlands that some control of soil and root-dwelling nematodes is given.

• Break crops including brassicas (mustard and oilseed rape), *Tagetes* and *Sorghum sudanense* (Sudan grass) are being researched in various countries, as a means of reducing soil-dwelling nematode populations before growing nematode-susceptible crops. These plants release various chemicals in the soil which have nematicidal activity. It is possible that the extraction and formulation of some of these chemicals may have potential as nematicides for control of LBN.

• Celery seed oil is commercially available as an essential oil and contains the compound sedanolide, which has been shown to have nematicidal activity, giving 100% mortality of free-living nematode species in laboratory tests (Momin & Nair, 2001).

If natural plant extracts proved to be effective against LBN, their compatibility with biological control agents used against other pests within IPM programmes is unknown and should be evaluated. For example, a recent study found that mustard green manures reduced the infection rates by beneficial entomopathogenic nematodes used in biological control programmes (Ramirez *et al.*, 2009).

### Host plant resistance

Resistant and tolerant cultivars against LBN have also been identified and could be developed as part of an Integrated Pest Management programme to reduce LBN infestation. Jagdale & Grewal (2006) identified four cultivars of *Hosta* spp. which were not infested by *A. fragariae*. In the UK, chrysanthemum varieties with resistance to *A. ritzemabosi* have been observed and these are described to be lacking a nutritional factor

which causes the infestation to be isolated and not spread to other leaves (Wallace, 1961). Despite this, the use of resistant cultivars remains limited in commercial nurseries partly due to the difficulties with breeding the resistance into acceptable cultivars (Roberts, 1992) and also to the lack of research and breeding effort with available germplasms unscreened for nematode resistance (Starr *et al.*, 2002).

In conclusion this review has determined that high standards of nursery and crop hygiene together with use of sub-irrigation remains the most effective cultural control measure against LBN. Vydate 10G is currently the only effective nematicide available in the UK for control of this pest but the review identified potential chemicals and plant extracts which could be evaluated.

# Conclusions

- The nematode products Nemasys L and SuperNemos significantly reduced numbers of live vine weevil larvae in substrate-grown strawberry when compared with untreated controls. Nemasys L and SuperNemos were equally effective. Vine weevil larvae feeding damage did not significantly affect plant vigour during the experiment period
- Percentage aphid hyperparasitism was between 17 and 70% on a HNS nursery during 2012. The hyperparasitoid species *Dendrocerus* sp. *Asaphes* sp. and *Alloystra brevis* were identified.
- During monitoring of an outdoor organic lettuce crop where *A. colemani* was regularly released, only a low proportion of aphids were parasitised. Live aphid numbers declined during monitoring which was likely to be due to natural infection by entomopathogenic fungi. Mummies which were collected from the crop confirmed the presence of hyperparasitism by several hyperparasitoid species.
- High standards of nursery and crop hygiene remain the most effective cultural control measure against leaf and bud nematode. Hot water treatments may also be effective but the efficacy and plant safety requirements for all susceptible HNS species and cultivars is not available. Currently Vydate 10G is the only effective nematicide for the control of the pest but the review identified other potential chemicals and alternative control methods that could justify evaluation.

# Knowledge and Technology Transfer

The results of each research project were discussed informally with the growers hosting the experiments and with the suppliers of the products tested. Specific knowledge transfer outputs from this Fellowship were:

Publications (with input from experienced ADAS colleagues):

HDC News article April 2013 (Gemma Hough and Jude Bennison).

Presentations:

Summary of the Fellowship project at ADAS Technical Skills Meeting April 2013 (Gemma Hough)

Scientific Conferences:

Royal Entomological Society Meeting July 2012- Insects in a human dominated world- The Horticultural Fellowship explained - Summary of the Fellowship project (Gemma Gillies).

AAB Advances in Biological Control meeting October 2012; The ADAS IPM Horticultural Fellowship (Gemma Gillies).

# Glossary

Hyperparasitism – when a primary parasitoid developing within its host is attacked by a secondary parasitoid. Here, this refers to naturally occurring hyperparasitoid species which attack the aphid parasitoids being used as biological control agents to control aphid pests.

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