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Authentication

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

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PRACTICAL SECTION FOR GROWERS

Commercial benefits of the project

The project has:

- Determined the distribution and scale of damage caused by capsid bugs in UK protected salad crops. Losses of £0.3 to £2 per m^2 have been recorded for cucumber crops.
- Made growers aware of damage caused by capsids that had previously been attributed to other factors, see HDC factsheet 37/96.
- Demonstrated the existence of a sex attractant produced by the female *Lygus rugulipennis* capsid that may be developed for use in baited monitoring traps.
- Improved the knowledge of the biology and behaviour of the two most important species of capsids, thus aiding the development of integrated control strategies.
- Identified an anti-feedant chemical (pymetrozine) that can be used within integrated pest management (IPM) programmes to reduce capsid damage to commercially acceptable levels. Pymetrozine as the product Chess, has on-label approval for use on protected cucumbers and specific off-label approval for use on protected peppers and aubergines.
- Identified an effective biological control agent (*Beauveria bassiana*) that can be used to control capsid bugs in pesticide-free production systems (subject to registration in the UK), and determined its compatibility with other crop protection agents.

Background and objectives

Capsids belong to the Miridae, a large family of small to medium sized soft-bodied insects that exploit a wide range of habitats. For many years, a plant feeding species, *Lygus rugulipennis* (European tarnished plant bug), has been a sporadic pest in U.K. glasshouses but until recently it has been uncommon for specific action to be taken against it in edible crops. When pest control was based entirely on chemicals, invading *L. rugulipennis* did not establish breeding populations on plants because they were killed by insecticides applied against other pests, such as *Trialeurodes vaporariorum* (glasshouse whitefly). With the advent of IPM in protected salad crops in the 1970s and 1980s, capsids were able to survive and colonise plants. However, reports of serious damage remained rare until the early-1990s, when the number of requests for advice relating to control of capsids began to increase markedly.

In 1996, the HDC commissioned a project aimed at improving the knowledge of the behaviour of plant feeding capsids in protected salads as a first step in formulating a sustainable control strategy within the existing IPM programmes. In summary, the specific objectives were:

- To improve the knowledge of the species involved, their natural habitats and the timing of crop invasion.
- To develop methods of monitoring the invasion of protected edible crops by capsids.
- To develop methods of controlling capsid bugs within IPM programmes in protected edible crops.

Summary of results and conclusions

Capsids – identifying damage symptoms and economic importance

The work in the first year of the project concentrated on determining the importance of capsid infestations in protected edible crops. Detailed crop monitoring at several sites in Yorkshire showed that two species were causing damage: *Lygus rugulipennis* in cucumber crops and *Liocoris tripustulatus* in pepper and aubergine crops.

The following symptoms were seen in cucumbers, peppers and aubergines:

i) feeding in growing points resulted in distorted growth and holes in leaves as they expanded;
ii) probing in stems just below growing points resulted in death of the growing point;
iii) feeding and egg laying in very young fruit resulted in distortion as the fruit swelled;
iv) feeding on maturing fruit resulted in surface scars which rendered the fruit unmarketable.

No damage was seen in tomato crops even where they were grown adjacent to heavily infested cucumber and pepper crops.

Based on the detailed monitoring information, an illustrated factsheet was produced to aid growers to recognise capsids and symptoms of their damage (HDC Factsheet 37/96) and this was circulated with a questionnaire to all cucumber, pepper and aubergine growers registered with the HDC. A total of 120 cucumber, pepper and aubergine growers responded to the survey and 48 confirmed that they had seen capsid activity in their crops. Approximately one third of those who had seen damage, reported that it occurred over large areas of their crops. The most seriously affected were cucumbers where the estimated financial loss ranged from £0.3 to £2 per m^2. Many of the growers who reported less serious damage had restricted the development of the problem by applying broad spectrum insecticides but indicated that this had affected biological control of other pests and had resulted in secondary problems. The survey confirmed that this was a nationwide problem that had become much more serious during the last four years.

Biology and behaviour of capsid bugs

Ms Fiona Hunter began a PhD at the University of Newcastle in 1998, aimed at improving the knowledge of natural habitats and activity patterns of *L. rugulipennis* and *L. tripustulatus*. The studies were focused on the insects' overwintering sites, spring migration, population dynamics and host plant choice. Laboratory studies have provided valuable background information about the development times of all life cycle stages of *L. rugulipennis*, and their activity patterns in natural habitats in northern England. Plant host choice experiments indicated that female *L. rugulipennis* preferred cucumber plants to nettle, which is their common host in the wild, and appeared to respond to chemical cues produced by the plants. The salad hosts contained more nitrogen than the wild plants and this may have been an important factor in the insect's

choice. Males did not show this preference. Both female and male *L. tripustulatus* exhibited a preference for nettle over pepper, and this appeared to be a response to visual rather than chemical cues.

Further details on this work may be obtained by contacting Fiona Hunter at the University of Newcastle.

Developing an integrated control strategy for capsids

Monitoring pest invasions

A reliable method of monitoring the migration of capsid bugs into greenhouses would enable control measures to be timed more accurately. Flat and cylindrical white and yellow traps were evaluated in cucumber and pepper crops that were infested with *L. rugulipennis* and *L. tripustulatus* respectively. The numbers of *L. rugulipennis* and *L. tripustulatus* caught were quite small, indicating that the insects were not specifically attracted to any of them. However, the numbers of male *L. rugulipennis* caught were greatly increased when the traps were baited with live females. Volatile chemicals, assumed to be sex pheromones, have been isolated from female *L. rugulipennis* and have potential as attractants in monitoring traps.

In 2001, traps baited with lures containing four different formulations of synthetic pheromones were evaluated against *L. rugulipennis* in a total of 14 commercial cucumber crops. There was no indication that the lures increased the attractiveness of the traps. This may have been because the dose and/or ratio of the chemical components were incorrect. Further R&D is required to develop pheromone baited traps for capsid bugs.

Evaluation of IPM compatible entomopathogenic fungi for the control of capsids

Three commercially available entomopathogenic fungi (*ie* two strains of *Verticillium lecanii* [Mycotal and Vertalec] and one *Beauveria bassiana* [Naturalis-L]) were evaluated against both *L. rugulipennis* and *L. tripustulatus* in laboratory bioassays in 1998. Naturalis showed greatest effect and was subsequently evaluated against *L. rugilipennis* on cucumber plants in experimental glasshouses. A single application of a high volume spray or a low volume mist provided similar results with numbers of adult *L. rugulipennis* being reduced by 60% compared to untreated controls. However, the relative humidity (RH) in the glasshouse was high (approximately 90% in the crop canopy) during that experiment, which favoured the fungal entomopathogen, and it was repeated in 1999 under more challenging conditions of lower RH (approximately 70%).

The second experiment (at 70% RH) was enlarged to incorporate an additional *B. bassiana* product (BotaniGard WP) that contained larger numbers of viable spores than Naturalis. Both products were applied three times at five day intervals by high volume spray or low volume mist. The overall mean number of *L. rugulipennis* in the *B. bassiana* treatments was 78% lower than the untreated control. The additional control compared to the first experiment was attributed to the three spray programme. The results suggested that infection of *L. rugulipennis* by *B. bassiana* was not dependant on high RH in the aerial environment.

Beauveria bassiana is active against a wide range of invertebrates and it is important to know the impact that it will have on the biological control agents used to control other pests in IPM programmes. The effects of the entomopathogen on the predatory mites, *Amblyseius cucumeris* and *Phytoseiulus persimilis*, and the parasitic wasps, *Encarsia formosa* and *Aphidius colemani*, were determined in laboratory-based bioassays between 1998 and 2000. There was no evidence to suggest that *B. bassiana* was harmful to *A. cucumeris* or *A. colemani.* Although it was demonstrated that the entomopathogen could infect *P. persimilis*, this did not have a significant effect on the population tested. However, *B. bassiana* was harmful to *E. formosa.* The results of two experiments demonstrated that *B. bassiana* sprayed on scales of *Trialeurodes vaporariorum* (glasshouse whitefly) parasitised by *E. formosa*, before and after they turned black, reduced survival of the parasitoid by 36% and 62% respectively. *Beauveria bassiana* also infects glasshouse whitefly directly and can provide effective control of this pest, the overall effect on this component of the IPM programme may not be detrimental.

The results of studies to determine the compatibility of *B. bassiana* with fungicides commonly used in UK cucumber and tomato crops indicated that Nimrod, Fungaflor and Scala were relatively safe and could be used within the same IPM programme. Rovral and Thiovit had already been shown to be safe in a previous project. However, the results indicated that Repulse and Amistar remained harmful to *B. bassiana* for at least seven days after application.

Evaluation of IPM compatible chemical insecticides for the control of capsids

The efficacy of two selective insecticides, buprofezin (as Applaud) and pymetrozine (as Chess), against *L. rugulipennis* was determined in small-scale experiments in 1998. There was no evidence to suggest that the insect growth regulator, buprofezin, prevented *L. rugulipennis* nymphs moulting or that it reduced oviposition by adult female *L. rugulipennis*. Although the antifeedant chemical, pymetrozine, did not kill adult *L. rugulipennis*, the damage to growing points of treated plants was much reduced compared to untreated controls in both laboratory bioassays and small scale experiments on plants.

Pymetrozine was further evaluated against *L. rugulipennis* on cucumber plants in experimental and commercial glasshouses in the final year of the project. Programmes of up to three applications of high volume sprays or low volume mists of pymetrozine (Chess) were evaluated against *L. rugulipennis* on cucumber plants in experimental glasshouses. The damage in the untreated control increased steadily over the four-week experimental period. Both HV and LV applications of pymetrozine prevented damage during the week following the first application of the anti-feedant chemical. Where there were no further applications, the damage increased steadily throughout the remainder of the experiment. Where HV or LV applications of pymetrozine were continued at weekly intervals, damage was restricted to commercially acceptable levels. Further evaluations of pymetrozine in commercial crops confirmed that *L. rugulipennis* damage to young cucumber plants (ie less than 1m high) could be prevented for 7 to 10 days when applied as a high volume spray at 400g per 1000 litres of water per hectare. ULV applications were also successful at 800g product per hectare (normally in 10 litres water per hectare). There was no evidence of any phytotoxic effects even when the

product was applied ULV to very young plants one day after planting in the glasshouse. A small scale glasshouse experiment demonstrated that pymetrozine (as Chess) also had the potential to prevent damage by *L. tripustulatus* to pepper crops. This is consistent with more recent observations in commercial pepper crops.

Action Points for Growers

- Capsids are difficult to detect at low population densities and the appearance of damaged growing points and fruit is often the first indication of their presence. Growers of cucumber, pepper and aubergine crops must be vigilant from May to the end of September and be prepared to act promptly to control the pest. Please refer to HDC factsheet 37/96 for identification of damage symptoms. As yet there are no traps developed to accurately monitor for the invasion of capsids. Pheromone-baited traps offer some potential but further studies are needed to develop monitoring traps for commercial use.

- Cucumber plants trained by the cordon system are very vulnerable to damage by capsids (*Lygus rugulipennis*) until they reach the support wire and are allowed to produce lateral shoots. Such damage may be reduced significantly by applying Chess (pymetrozine) HV at 400g of product per 1000 litres of water to maximum leaf retention, or ULV at 800g of product per hectare (normally in 10 litres of water). ***Growers should note that the maximum application rate for either application method is 800g Chess per hectare.***

- To provide continuous protection of cucumber crops throughout periods of sustained capsid activity, applications of Chess are required at weekly intervals. Growers should note that the total quantity allowed is 2.4 kg of product per hectare per crop.

- Small-scale experiments have indicated that Chess will provide pepper plants with protection from capsids (*Liocoris tripustulatus)*. Use Chess on peppers is allowed under specific off-label approval.

- Laboratory and crop scale experiments demonstrated the potential of the fungal entomopathogen, *Beauveria bassiana*, as Naturalis-L, for the control of capsid bugs in cucumbers and peppers. ***However, the results from this work cannot be exploited yet as the product is not approved for use in the UK.***

Note on pesticide approvals

Chess (pymetrozine) has on-label approval for use on protected cucumber crops. This approval was granted in the UK in 2000. The statutory regulations on the label state a maximum total dose of 2.4 kg Chess per hectare to each crop, with a maximum individual dose of 0.8 kg Chess per ha. A minimum spray interval of one week is required between applications.

Chess (pymetrozine) has specific off-label approval for use on protected crops of pepper and aubergine; SOLAs 2337/2000 and 1683/2001. The SOLA specifies a

maximum individual dose of 0.8 kg Chess per 100 litres water and a maximum number of treatments of 3 per crop.

Naturalis-L and BotaniGard (both contain *Beauveria bassiana*) are not currently registered for use in the UK.

Anticipated practical and financial benefits from the study

The provision of robust, sustainable and manageable strategies for the control of capsids in cucumber, pepper and aubergine crops will:

- Avoid direct damage and financial losses caused by these pests.
- Avoid secondary problems associated with the breakdown of IPM in these crops.
- Help to satisfy demands of UK's leading food retailers for produce grown under minimal pesticide regimes.

It is anticipated that ornamental crops will also suffer damage from capsids as growers move towards full IPM strategies. Therefore, the acquisition of knowledge and the development of new control measures against these pests will ultimately provide greater benefits within the whole UK horticultural industry.

SCIENCE SECTION

INTRODUCTION

Background

Capsids belong to the Miridae, a large family of small to medium sized soft-bodied insects that exploit a wide range of habitats. For many years, a plant feeding species, *Lygus rugulipennis* (European tarnished plant bug), has been a sporadic pest in UK glasshouses (Wardlow, 1985) but until recently it has been uncommon for specific action to be taken against it in edible crops. When pest control was based entirely on chemicals, invading *L. rugulipennis* did not establish breeding populations on plants because they were killed by insecticides applied against other pests, such as *Trialeurodes vaporariorum* (glasshouse whitefly). With the advent of IPM in protected salad crops in the 1970s and 80s, capsids were able to survive and colonise plants. However, reports of serious damage remained rare until the early-1990s, when the number of requests for advice relating to control of capsids began to increase markedly. In 1996, the Horticultural Development Council (HDC) commissioned a project aimed at improving the knowledge of the behaviour of plant feeding capsids in protected salads as a first step in formulating a sustainable control strategy within the existing IPM programmes.

Scientific/technical targets of the project

Years 1 and 2

1. A literature search to ensure that the research team has all available information.
2. The pest's activity will be monitored at selected sites to improve the knowledge of the species involved, their natural habitats and the timing of crop invasion.
3. A fact sheet will be prepared aimed at improving grower awareness of the damage caused by capsids.
4. An industry survey will be completed to determine the full extent of the problem in protected edible crops.

Years 3 to 5

In 1998, the project was extended to encompass the following additional targets:

1. To improve the knowledge of the biology and behaviour of *L. rugulipennis* and *L. tripustulatus*, with particular reference to their activity in protected edible crops.
2. To develop methods of monitoring the invasion of protected edible crops by capsids.
3. To develop methods of controlling *L. rugulipennis* and *L. tripustulatus* within IPM programmes in protected edible crops.
4. To improve the knowledge of the natural habitats and activity patterns of *L. rugulipennis* and *L. tripustulatus*.

Summary of work completed in Years 1, 2, 3 and 4 (Jacobson, 1997; Jacobson, 1998; Jacobson, 1999; Jacobson, 2000)

During 1996 and 1997, cucumber, tomato, pepper and aubergine crops were monitored at sites that had suffered intermittent damage by capsids in recent seasons. Two species of capsids were found to be causing damage; *Lygus rugulipennis* in cucumbers and *Liocoris tripustulatus* in peppers and aubergines. No damage was seen in tomato crops even where they were grown adjacent to heavily infested cucumber and pepper crops. The following symptoms were seen in cucumbers, peppers and aubergines:

i) capsid feeding in growing points resulting in distorted growth and holes in leaves as they expanded
ii) capsid probing in the stem just below the growing point resulting in death of the growing point
iii) capsid feeding and egg laying in very young fruit resulting in distortion as the fruit swelled
iv) capsid feeding on maturing fruit resulting in surface scars which rendered the fruit unmarketable.

Based on the detailed monitoring information, an illustrated fact sheet was produced to aid growers recognise capsids and symptoms of their damage (Jacobson & Hargreaves, 1996). In March 1997, this fact sheet was circulated with a questionnaire to all cucumber, pepper and aubergine growers registered with the HDC. Of 120 growers who responded to the survey, 48 confirmed that they had seen capsid activity and approximately one third of those reported that it occurred over large areas of their crops. The most seriously affected were cucumbers where the estimated financial loss ranged from £0.3 to £2 per m^2. Many growers who reported less serious damage had restricted the development of capsid infestations by applying insecticides but indicated that this disrupted IPM and resulted in secondary problems with other pests. The reported occurrences of *L. rugulipennis* in cucumber crops indicated that there was a small invasion in late-spring followed by a much larger invasion in July. This is consistent with the described bivoltine life cycle (Southwood, 1956; Easterbrook, 1997). The activity of *L. tripustulatus* was more evenly spread between May and September. Approximately 90% of growers who reported damage by capsids, had first seen it during the last five years; thus confirming that infestations were becoming more common in protected salad crops. There were no distinct differences between geographical regions showing this to be a national problem. Detailed examination of vegetation in the immediate vicinity of infested glasshouses failed to identify important breeding sites, so it must be assumed that both species migrate to the glasshouses from other locations. It is unclear why capsids should have become more troublesome in cucumbers, peppers and aubergines in recent years. It is unlikely that the behaviour of the two species has simultaneously changed, so it is probably due to differences in the availability of plant food or natural habitats. As there has been no widespread change in crop husbandry or insecticide usage that could have allowed capsids a new opportunity to colonise these crops, the most probable explanation is a change in their natural habitats outside the glasshouses leading to larger invasions.

A reliable method of monitoring the migration of capsid bugs into greenhouses would enable control measures to be timed more accurately. In 1996, flat and cylindrical white

and yellow traps were evaluated in cucumber and pepper crops that were infested with *L. rugulipennis* and *L. tripustulatus* respectively. The numbers of *L. rugulipennis* and *L. tripustulatus* caught were quite small, indicating that the insects were not specifically attracted to any of them. However, the numbers of male *L. rugulipennis* caught were greatly increased when the traps were baited with live conspecific females. Volatile chemicals, assumed to be sex pheromones, were subsequently isolated from female *L. rugulipennis* (Innocenzi *et al*., 1998). A preliminary evaluation of prototype pheromone baited traps in 1998 did not provide conclusive results, possibly because the dose and/or ratio of the component chemicals were incorrect. Modified lures were not available in 1999 or 2000 due to technical difficulties with their production at NRI but were tested in the final year of the project.

Three commercially available entomopathogenic fungi (*i.e*. two strains of *Verticillium lecanii* [Mycotal and Vertalec] and one *Beauveria bassiana* [Naturalis]) were evaluated against both *L. rugulipennis* and *L. tripustulatus* in laboratory bioassays in 1998. Naturalis showed greatest effect and was subsequently evaluated against *L. rugilipennis* on cucumber plants in experimental glasshouses. A single application of a high volume spray or a low volume mist provided similar results with numbers of adult *L. rugulipennis* being reduced by 60% compared to untreated controls. However, the relative humidity (RH) in the glasshouse was high (approximately 90% in the crop canopy) during that experiment, which favoured the fungal entomopathogen, and it was repeated in 1999 under more challenging conditions of lower RH (approximately 70%). The second experiment was enlarged to incorporate an additional *B. bassiana* product (BotaniGard WP) that contained larger numbers of viable spores than Naturalis. Both products were applied three times at five day intervals by high volume spray or low volume mist. The overall mean number of *L. rugulipennis* in the *B. bassiana* treatments was 78% lower than the untreated control. The additional control compared to the first experiment was attributed to the three spray programme. The results suggested that infection of *L. rugulipennis* by *B. bassiana* was not dependant on high RH in the aerial environment.

Beauveria bassiana is active against a wide range of invertebrates and it is important to know the impact that it will have on the biological control agents used to control other pests in IPM programmes. The effects of the entomopathogen on the predatory mites, *Amblyseius cucumeris* and *Phytoseiulus persimilis*, and the parasitic wasps, *Encarsia formosa* and *Aphidius colemani*, were determined in laboratory-based bioassays between 1998 and 2000. There was no evidence to suggest that *B. bassiana* was harmful to *A. cucumeris* or *A. colemani.* Although it was demonstrated that the entomopathogen could infect *P. persimilis*, this did not have a significant effect on the population tested. However, *B. bassiana* was harmful to *E. formosa.* The results of two experiments demonstrated that *B. bassiana* sprayed on parasitised *Trialeurodes vaporariorum* (glasshouse whitefly) scales, before and after they turned black, reduced survival of the parasitoid by 36% and 62% respectively. *Beauveria bassiana* also infects *T. vaporariorum* directly and can provide effective control of this pest, so the overall effect on this component of the IPM programme may not be detrimental.

The efficacy of two selective insecticides, buprofezin and pymetrozine, against *L. rugulipennis* was determined in small-scale experiments in 1998. There was no evidence to suggest that the insect growth regulator, buprofezin, prevented

L. rugulipennis nymphs moulting or that it reduced oviposition by adult female *L. rugulipennis*. Although the antifeedant chemical, pymetrozine, did not kill adult *L. rugulipennis*, the damage to growing points of treated plants was much reduced compared to untreated controls in both laboratory bioassays and small scale experiments on plants. Pymetrozine was further evaluated against *L. rugulipennis* on cucumber plants in experimental and commercial glasshouses in the final year of the project.

The ecology of *L. rugulipennis* and *L. tripustulatus* is not well known. Ms Fiona Hunter started a PhD in 1998, aimed at improving the knowledge of natural habitats and activity patterns of *L. rugulipennis* and *L. tripustulatus*. The studies have focused on the insects' overwintering sites, spring migration, population dynamics and host plant choice. Much of the work prior to April 2000 was devoted to developing and testing experimental methods.

Year 5 (2000/2001) – Work plan

Following the Project Review Meeting in March 2000, the following work plan was agreed for years 4/5 of the project:

1. To evaluate traps baited with sex pheromones for monitoring the migration of *L. rugulipennis* into protected edible crops.
2. To determine the efficacy of pymetrozine (Chess) against *L. rugulipennis* on artificially infested experimental cucumber crops and naturally infested commercial cucumber crops.
3. To evaluate the potential of pymetrozine (Chess) to prevent *L. tripustulatus* causing damage to pepper plants.
4. To determine the compatibility of *B. bassiana* with fungicides commonly used in UK cucumber and tomato crops.
5. To improve the knowledge of natural habitats and activity patterns of *L. rugulipennis* and *L. tripustulatus.*

PART 1 – DEVELOPMENT OF METHODS OF MONITORING CAPSID INVASION

Experiment title:

To monitor the efficacy of pheromone traps against *Lygus rugulipennis* on commercial cucumber nurseries.

Introduction:

Capsid bugs may invade protected crops as early as April but the main migration occurs when populations increase in their natural habitats during the summer. Despite their relatively large size, capsids can be difficult to find in glasshouses and the appearance of damaged growing points and fruit is often the first indication of their presence. Improved monitoring procedures are required to detect capsid invasion and indicate when control measures should be applied.

Although standard white and yellow sticky traps were not very attractive to *L. rugulipennis*, the numbers of males caught were greatly increased when the traps were baited with live conspecific females (Jacobson, 1998). Volatile chemicals, assumed to be sex pheromones, were isolated from female *L. rugulipennis* and incorporated in lures for use in monitoring traps (Innocenzi *et al*., 1998). A preliminary evaluation of prototype pheromone traps in capsid infested cucumber crops during 1998 did not provide conclusive results.

Modified lures were not available in 1999 or 2000 due to technical problems with their production at NRI but they were evaluated in commercial crops during the 2001 growing season.

Materials and methods:

Trap design: Each *Lygus rugulipennis* trap consisted of a flat yellow sticky trap with two holes drilled close to the center in which were placed the cylindrical polythene vials (25mm x 8mm dia.) containing the lures. Lures were changed every four weeks.

Components in lures: The chemical components of the lures were supplied in more than one vial to avoid possible interactions:

Vial 1 - 100g (E)-2-hexenyl butyrate
Vial 2 - 100g (E)-2-hexenyl butyrate + hexyl butyrate 2:3
Vial 3 - 10mg (E)-4-oxo-2-hexenal
Vial 4 - 100g of 10% (E)-2-hexenyl butyrate in polyethylene glycol
Vial 5 - 100g of 10% (E)-2-hexenyl butyrate + hexyl butyrate 2:3 in polyethylene glycol
Vial 6 - 20mg of 10% (E)-4-oxo-2-hexenal in polyethylene glycol

Treatments: Original lures used from 25 July 2001 to 6 September 2001:

T1. vial 2 plus vial 3
T2. vial 1 plus vial 3
T3. Untreated control – empty lure tubes

Modified lures used from 6 September 2001 to 27 September 2001:

T4. vial 5 plus vial 6
T5. vial 4 plus vial 6
T6. Untreated control – empty lure tubes

Site details: Sites were selected that met the following criteria:

- recently replanted cucumber crops
- a history of damaging infestations of *L. rugulipennis*
- sightings of *L. rugulipennis* in crops this season.

Site-glasshouse reference	Location (all East Yorkshire)	Approximate size of glasshouse (m^2)	Monitoring period with:	
			Original lures	Modified lures
1-1	Brough	4000	18/7-6/9	-
1-2		4000	18/7-6/9	-
2-1	Brough	4000	18/7-6/9	-
2-2		2000	-	6/9-27/9
2-3		1000	-	6/9-27/9
2-4		2000	-	6/9-27/9
2-5		3000	-	6/9-27/9
3-1	Everthorpe	2000	18/7-6/9	6/9-27/9
4-1	South Cave	10000	25/7-6/9	-
4-2		10000	25/7-6/9	-
5-1	South Cave	4000	25/7-12/9	-
5-2		4000	25/7-12/9	-
6-1	Welton	7200	18/7-6/9	6/9-27/9
6-2		4800	18/7-6/9	6/9-27/9

Trap position: One trap of each Treatment was placed in each glasshouse. The traps were spaced as far apart as possible and fastened to crop wires using bulldog clips. The positions of the traps within each glasshouse were changed at weekly intervals.

Assessments: Traps were examined at weekly intervals. The numbers of male and female *L. rugulipennis* were recorded. Dead insects were removed from the traps and they were repositioned.

Results and Discussion:

The total numbers of female and male *L. rugulipennis* caught on each type of trap at each site from 25 July to 6 September are shown in Table 2. There was no indication that the lures were attractive to either males or females. It was possible that the restricted ventilation in the glasshouses resulted in the atmosphere becoming saturated with the pheromones, so that the insects were unable to find the source of the chemicals. Therefore, from 6 September to 27 September, modified lures with reduced concentrations of pheromones were used but there was still no evidence of attraction (Table 3). This may be because the dose and / or ratio of the chemical components were incorrect and this is being further investigated in laboratory studies at NRI.

Overall, the traps with synthetic pheromones appeared to show some repellency, particularly for females, when compared to unbaited traps. It is therefore possible that some of the chemical components, or specific combinations of those components, have defensive properties. The results may be further complicated by *L. rugulipennis* captured on traps releasing some form of alarm pheromone that causes behavioural responses in other individuals. These factors are also being investigated further in laboratory studies at NRI.

The cumulative numbers of female and male *L. rugulipennis* caught on each type of trap at all sites between 27 July and 6 September 2001 are shown in Figures 1 and 2 respectively. These results do not show any periods of intensive *L. rugulipennis* activity. Very few males or females were caught after 6 September 2001 despite being quite easy to find on the plants at several sites.

Conclusion:

Although pheromone baited traps have not yet been successfully used to attract male or female *L. rugulipennis* in glasshouse or field crops, the ability to monitor the invasion of this species remains an important component of the overall control strategy.

Table 2. The total numbers of female and male *L. rugulipennis* caught at each site from 25 July to 6 September 2001.

Site	T1		T2		T3	
	males	females	males	females	Males	Females
1-1	0	2	0	1	0	2
1-2	0	0	0	1	1	1
2-1	0	0	0	0	0	1
3-1	0	2	2	3	5	5
4-1	0	1	0	0	0	0
4-2	1	1	1	0	2	5
5-1	0	1	1	3	4	12
5-2	3	0	4	3	2	0
6-1	1	1	0	1	5	4
6-2	1	2	2	0	2	4
TOTAL	6	10	10	12	21	34

Table 3. The total numbers of female and male *L. rugulipennis* caught at each site from 6 September to 27 September 2001.

Site	T4		T5		T6	
	males	females	males	females	Males	Females
2-2	0	0	0	0	0	0
2-3	0	0	0	0	0	0
2-4	0	0	0	0	0	0
2-5	0	0	0	0	0	0
3-1	1	0	0	1	0	0
6-1	0	0	0	0	0	0
6-2	0	0	1	0	0	0
TOTAL	1	0	1	1	0	0

Figure 1. Cumulative numbers of female *L. rugulipennis* on each type of trap at all sites between 27 July and 6 September 2001.

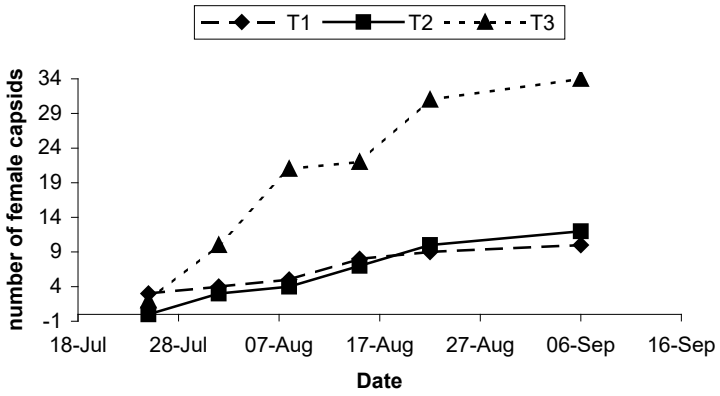
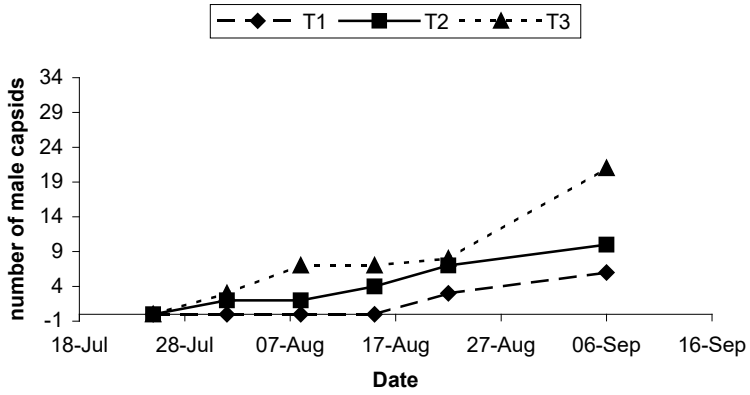


Figure 2. Cumulative numbers of male *L. rugulipennis* on each type of trap at all sites between 27 July and 6 September 2001.



PART 2 – CONTROL OF *LYGUS RUGULIPENNIS* WITH PYMETROZINE IN EXPERIMENTAL GLASSHOUSES

Experiment title:

To evaluate pymetrozine against *Lygus rugulipennis* on cucumber plants in an experimental glasshouse.

Introduction:

The antifeedant chemical, pymetrozine, (as Chess), is specific to some plant-sucking insects of the order Hemiptera. For example, following application of pymetrozine to plants, aphids are reported to die due to starvation within 4 days. The product is now registered for use on cucumbers in the U.K. and could be integrated in the IPM programme.

As capsid bugs also belong to the order Hemiptera, preliminary studies were done in the laboratory in 1998 and 1999 to determine whether pymetrozine caused mortality of *L. rugulipennis* directly or indirectly through antifeedant properties. Although the chemical did not kill adult *L. rugulipennis*, the damage to growing points of treated plants was less severe than untreated controls. This was further investigated in the present experiment.

Materials and Methods:

Site: HRI, Stockbridge House.

Glasshouse: Headley Hall Glasshouse Unit.

Treatments:
1. Untreated control.
2. Single pymetrozine (Chess) HV spray.
3. Three pymetrozine (Chess) HV sprays at 7 day intervals.
4. Single pymetrozine (Chess) LV mist.
5. Three pymetrozine (Chess) LV mist applications at 7 day intervals.

Rates of application:
HV Spray - 40g product per 100 litres water.
LV Mist - 40g product per 1000m^2.

Application: Pymetrozine was applied in Treatments 2 to 5 on 5 September 2000. The additional applications to Treatments 3 and 5 were done on 12 and 19 September 2000. High volume sprays were applied to maximum leaf retention, using a fully calibrated Oxford Precision Sprayer, at volumes equivalent to 720, 930 and 1125 litres per hectare on 5, 12 and 19 September respectively. The volumes used increased because the plants

were growing and retained more spray. The low volume mist treatments were applied using a fully calibrated Turbair 240 Scamp LV Mist sprayer at volume equivalent to 16.6 litres per hectare.

Plants: Cucumber, cv Enigma.

Planting date: 30 August 2000.

Growing medium: The plants were grown hydroponically in peat bags with excess feed solution running to waste and were trained by the cordon-V system.

Experimental design: The experiment was designed after consultation with Dr Julie Jones (Biometrician, HRI, Wellesbourne). Due to the pest's mobility, each treatment was confined to a separate glasshouse section. Whole treatments were not replicated but each section contained twenty four plants arranged in four plots.

Pest infestation: A total of 750 adult *L. rugulipennis* were collected from natural habitats between 5 and 26 September 2000 and released in equal numbers in each of the glasshouse sections.

Assessment: Assessments were done in all Treatments immediately before each of the three applications of pymetrozine, and 7 days after the final application. On each occasion, the youngest 10cm of growth of every plant was examined and capsid damage recorded using the following index:

0	No visible damage.
1	< 2% damaged (ie "pin-prick" damage, tearing and/or distortion).
2	2-10% damaged.
3	10-40% damaged.
4	40-100% damaged.
5	Growing point killed.

Analysis of data: Analysis of variance was done on log transformed data (with the term 0.375 added to all data due to the large number of zeros) for Treatments 1, 2, 3 and 4 on separate and combined post-treatment assessment dates, and the differences were compared using LSD. Data for Treatment 5 could not be included in the analysis because all values were zero. Some caution is required when interpreting the results from experiments such as this where it is impractical to replicate treatments fully. There is no true replication and within treatment variation is used as a measure of experimental variation.

Results and Discussion:

The mean damage indices for each Treatment at each assessment date are shown in Table 4. For ease of interpretation, these means are also shown in Figure 3.

No damage was recorded before the first application of pymetrozine, which coincided with the first release of *L. rugulipennis*. Thereafter, there were some significant differences between Treatments. Over the full course of the experiment, there was more ($P<0.05$) damage in the untreated control than in any of the pymetrozine Treatments, and more ($P<0.05$) damage in the single HV spray and LV mist Treatments than in the multiple HV spray Treatment ($P<0.05$). No damage was recorded in the multiple LV mist Treatment.

On the first post-treatment assessment date (week 1), the mean damage index in the untreated control was 1.08, while there was no damage recorded in any of the pymetrozine Treatments. On the second post-treatment assessment, there was more ($P<0.05$) damage in the untreated control (mean index 0.88) than the 3 x LV and 3 x LV pymetrozine Treatments (mean indices 0 and 0.04 respectively). Neither the 1 x LV nor 1 x HV Treatments were significantly different to the untreated control at that time. The situation was similar on the third and fourth post-treatment assessments, with mean damage indices in the untreated controls, 3 x LV and 3 x HV Treatments being 1.42, 0 and 0.04 respectively on the third assessment and 1.71, 0 and 0.25 respectively on the fourth assessment.

The trends throughout the experiment can be seen clearly in Figure 3. The damage in the untreated control increased steadily over the four-week period. Both LV and HV applications of pymetrozine prevented damage during the week following the first application of the anti-feedant chemical. Where there were no further applications of pymetrozine (Treatment 2 [1 x LV] and Treatment 4 [1 x HV]), the damage increased steadily throughout the remainder of the experiment. Where HV spray applications of pymetrozine continued at weekly intervals, damage was restricted to commercially acceptable levels. The weekly applications of LV mists of pymetrozine completely prevented damage.

Conclusions:

These results indicate that a HV spray or LV mist application of pymetrozine will protect cucumber plants from damage by *L. rugulipennis* for between one and two weeks. To provide continuous protection throughout periods of sustained *L. rugulipennis* activity, applications would have to be repeated at weekly intervals.

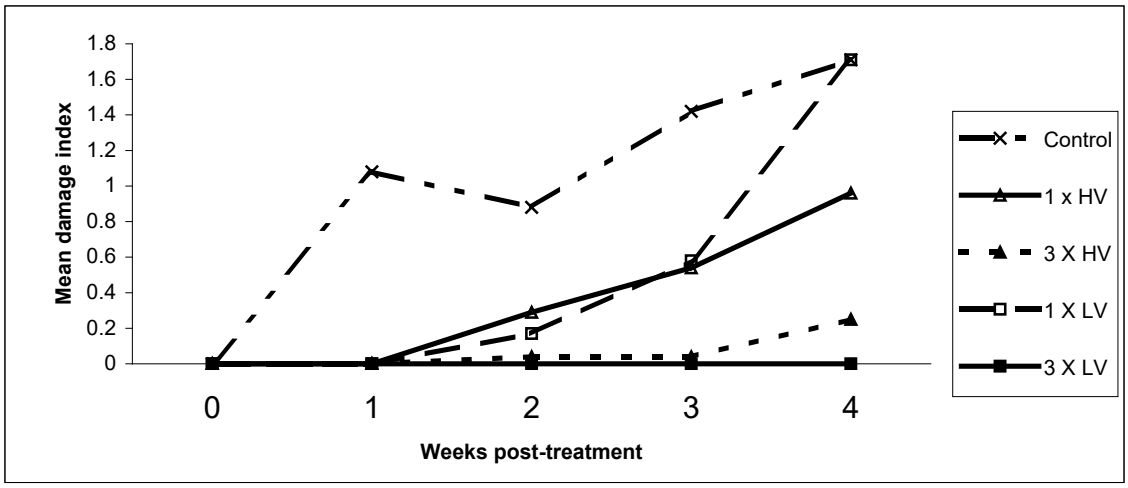
Table 4. Mean damage index (log transformed) in all Treatments immediately before the first application of pymetrozine and at weekly intervals over the following four weeks.

Assessment (weeks post-treatment)	Mean damage index (log transformed) on five assessment dates in Treatments (#) :					
	1. (Control)	2. (1 x HV)	3. (3 x HV)	4. (1 x LV)	5. (3 x LV)*	LSD [df =12]
0	0	0	0	0	0	-
1	1.08 (-0.22)	0 (-0.98)	0 (-0.98)	0 (-0.98)	0	- (0.48)
2	0.88 (-0.48)	0.29 (-0.76)	0.04 (-0.93)	0.17 (-0.76)	0	- (0.41)
3	1.42 (-0.04)	0.54 (-0.47)	0.04 (-0.93)	0.58 (-0.50)	0	- (0.74)
4	1.71 (0.18)	0.96 (-0.18)	0.25 (-0.77)	1.71 (0.20)	0	- (0.93)

* excluded from analysis because all figures were zero

for comparisons between Treatments 1, 2, 3 and 4 within assessment dates use the LSD [df] shown in table, and between assessment dates use LSD = (0.30) [df = 48].

Figure 3. Mean *Lygus rugulipennis* damage index for each treatment at each assessment.



PART 3 – EFFICACY OF PYMETROZINE AGAINST *LYGUS RUGULIPENNIS* IN COMMERCIAL CUCUMBER CROPS

Experiment title:

To evaluate HV and ULV applications of pymetrozine against *Lygus rugulipennis* on commercial cucumber crops.

Introduction:

Previous studies in small-scale experimental glasshouses indicated that applications of pymetrozine (Chess) could significantly reduce damage to young cucumber plants by *L. rugulipennis*. Members of the Cucumber Technology Group requested that both HV and ULV applications be evaluated on a larger scale in young commercial crops with vulnerable growing points. Ideally, such treatments would be evaluated in comparison to untreated controls but this proved difficult to organise because growers who had suffered serious capsid damage in the past were reluctant to leave their crops unprotected. The evaluations were, therefore, done by comparing damage before and after application of pymetrozine.

Materials and methods:

Sites and treatments: The work was done at four sites:

Site 1 was 1000m^2 of cucumber plants, cv Media, that were attacked by *L. rugulipennis* immediately after planting on 5 September 2001. At that stage the plants were approximately 0.4m high. Pymetrozine was applied on 6 September 2001 by ULV at a rate equivalent to 800g per 100 litres per hectare.

Site 2 was 2000m^2 of cucumber plants, cv Media, that were also attacked by *L. rugulipennis* immediately after planting on 5 September 2001. Pymetrozine was applied on 6 September 2001 by ULV at a rate equivalent to 800g per 50 litres per hectare.

Site 3 was 4000m^2 of cucumber plants, cv Enigma, that were approximately 1m high when *L. rugulipennis* activity was observed on 6 August 2001. The crop was sprayed high volume at a rate equivalent to 400g per 1000 litres per hectare on 6 August.

Site 4 was 3600m^2 of cucumber plants, cv Tiffany, that were approximately 0.6m high when *L. rugulipennis* activity was observed on 7 August 2001. The crop was sprayed high volume at a rate equivalent to 400g per 1000 litres per hectare on 7 August.

Assessments: The assessments varied slightly between sites because they had to take into account differences in the age of the crop, size of plants, crop layout and the amount of notice of treatments given by the individual growers.

At **Sites 1 and 2**, the pre- and post-treatment assessments were done on 6 and 13 September 2001 respectively on blocks of 558 plants (6 rows of 93) and 522 plants (6 rows of 87) respectively. On each occasion, the youngest 10cm of growth of every plant was examined and capsid damage was recorded using the damage index described in Part 2 of this report.

At **Sites 3 and 4**, single assessments were done on 15 August 2001, on blocks of 532 plants (4 rows of 133) and 516 (4 rows of 129) plants respectively. The youngest 10cm of growth of every plant was examined and capsid damage was recorded using the damage index described in Part 2 of this report. In addition, the leaves positioned 8-12 leaves below the growing point were examined as these were at the top of the plant when the treatments were applied and showed a historical record of the damage that was present at that time.

Analysis of data: Formal analysis of data was not possible.

Results and Discussion:

At **Site 1**, 47 of 522 plants examined before application of pymetrozine showed some damage by *L. rugulipennis*, including 8 plants with destroyed growing points. The mean index on the damaged plants was 2.4 and the mean index overall was 0.2. The post-treatment assessment revealed an overall mean index of 0.03.

84 of 558 plants at **Site 2** showed some damage before treatment, including 10 with destroyed growing points. The mean index on the damaged plants was 2.1 and the mean index overall was 0.3. Damage was reduced to a mean index of 0.01 at the post-treatment assessment.

The pre-treatment damage was less severe at **Sites 3 and 4**, with overall mean indices of 0.05 and 0.04 respectively. The post-treatment damage was negligible (indices <0.002) at both sites.

If there had been no constraint on *L. rugulipennis* feeding, the damage would have been expected to continue and be greater at the end than at the start of the experimental period. In fact, the damage decreased at all sites following application of pymetrozine regardless of the method of application.

The results indicate that pymetrozine (Chess) will prevent damage by *L. rugulipennis* to young cucumber plants (ie less than 1m high) for 7 to 10 days when applied HV at 400g per 1000 litres of water per hectare, or ULV at 800g product per hectare (normally in 10 litres water). These findings are consistent with the previous results from more detailed studies in small-scale experimental glasshouses that tested HV spray and LV mist applications.

There was no evidence of any phytotoxic effects even when the product was applied ULV to very young plants one day after planting in the glasshouse.

PART 4: POTENTIAL OF PYMETROZINE AGAINST *LIOCORIS TRIPUSTULATUS* ON PEPPER PLANTS

Experiment title:

A preliminary investigation of the effect of pymetrozine against *Liocoris tripustulatus* on pepper plants.

Introduction:

Previous laboratory and small-scale glasshouse experiments had demonstrated that the anti-feedant chemical, pymetrozine, could significantly reduce feeding damage by *Lygus rugulipennis* on cucumber plants. The present experiment was designed to indicate whether this chemical also had the potential to reduce feeding damage by *Liocoris tripustulatus* on pepper plants.

Materials and Methods:

Site: HRI, Stockbridge House, Glasshouses FF 5 and 9.

Treatments:
1. Untreated control – *L. tripustulatus* on pepper plants sprayed with water.
2. *L. tripustulatus* on pepper plants that had been sprayed with pymetrozine (Chess) at the rate equivalent to 40g per 100l water.

Application: The treatments were applied on 28 September 2000 when the plants were approximately 0.6m high. The sprays were applied high volume to maximum leaf retention using a fully calibrated Oxford Precision Sprayer.

Plants: Pepper, cv Mazurka.

Planting date: 23 August 2000.

Growing conditions: The plants were grown hydroponically in rockwool slabs with excess feed solution running to waste. Two shoots were allowed to develop on each plant and they were trained up separate vertical strings attached to a support wire approximately 2m above ground. The glasshouse environment was consistent with normal commercial pepper production.

Experimental design: Due to the mobility of the pest, the two Treatments were housed in separate glasshouses. There was one plant shoot per replicate and twelve replicates per Treatment.

Pest infestation: *L. tripustulatus* were reared in culture at HRI Stockbridge House. A total of 66 adult females were released in equal numbers in the two glasshouses on three occasions between 28 September and 3 October 2000.

Assessment: Assessments were done immediately before application of pymetrozine, and 7 and 12 days post-application of pymetrozine. On each occasion, the youngest 6cm of growth (ie growing point and approximately 3 youngest leaves) was examined and *L. tripustulatus* damage recorded using the following index:

Score	Damage
0	No visible damage.
1	2% damaged (ie "pin-prick" damage, tearing and/or distortion).
2	2-10% damaged.
3	10-40% damaged
4	40-100% damaged
5	Growing point killed.

Analysis of data: Formal analysis of the data was not possible.

Results and Discussion:

The mean and range of the damage index in each treatment at each assessment date are shown in Table 5. There was no damage before the treatments were applied. Although formal statistical analysis of these data was not possible, there was an apparent difference between the two Treatments. While the damage to pymetrozine treated plants was acceptable, the damage to untreated plants would probably have resulted in financial loss.

Table 5. Mean (range) of damage index in each treatment at each assessment date.

Treatment	Mean (range) damage index		
	Immediately before application	7 days post-application	12 days post-application
Untreated control	0	2.4 (1-5)	3.0 (2-5)
Pymetrozine	0	0.75 (0-2)	0.2 (0-1)

Conclusion:

Pymetrozine has the potential to prevent damage by *L. tripustulatus* to pepper crops. These observations should be confirmed in larger scale experiments in experimental or commercial glasshouses.

PART 5 – COMPATABILITY OF *BEAUVERIA BASSIANA* WITH FUNGICIDES COMMONLY USED IN UK CUCUMBERS AND TOMATOES

Experiment title:

To determine the compatibility of *Beauveria bassiana* with five fungicides commonly used in UK cucumber and tomato crops.

Introduction:

To integrate *B. bassiana* successfully into protected salad IPM programmes, it is essential to determine the compatibility of the fungal pathogen with the fungicides commonly used against the diseases of these crops.

In previous studies (Jacobson, 2000a), a bioassay was developed and used to determine the effect of seven fungicides on the germination of *B. bassiana* spores. Rovral and Thiovit were relatively safe at their recommended application rates. Scala was safe and Nimrod was relatively safe at one tenth of their recommended rates. Fungaflor, Repulse and Amistar prevented germination at one tenth of their recommended rates. The present experiment investigated the persistence of the effects of the five fungicides that had previously been shown to be harmful to *B. bassiana* at their recommended application rates.

Materials and Methods:

Site: HRI, Stockbridge House.

Treatments:

Product	Active ingredient	Formulation	% a.i	Rate (On or Off-Label)	a.i.(g) per l of media
Nimrod	Bupirimate	250g/l EC	25	200ml per 100l	0.5
Fungaflor	Imazalil	200g/l EC	20	50ml per 100l	0.1
Repulse	Chlorothalonil	500g/l SC	50	220ml per 100l	1.1
Amistar	Azoxystrobin	250g/l SC	25	80ml per 100l	0.2
Scala	Pyrimethanil	400g/l SC	40	100ml per 100l	0.4

Expt design: Five fungicides were tested at their recommended application rates immediately after treatment and following storage for 1, 3 and 7 days.

There was also one untreated control, giving a total of 24 treatments. There were five replicates of each treatment.

Preparation of replicates: 10 ml aliquots of molten malt extract agar (MEA) were poured into 50mm diameter single vent Petri dishes and allowed to solidify. Each dish formed one replicate. Each replicate was divided into three sections by marking the underside of the dish and a small circle (approx. 2 cm diameter) was drawn within each of the three sections. A Potter Tower (Potter, 1952) was used to apply 2ml of the recommended dilution of each fungicide (or water in the untreated control) at 0.5 bar to the surface of the MEA. The treated dishes were then stored under glasshouse conditions (20-25 ºC; 16 hour day including supplementary light) for the appropriate time.

Spore innoculation: A suspension of *B. bassiana* spores (BotaniGard WP at 125g per 100l water) was prepared immediately before use and the dilution adjusted with sterile 0.01% Tween 80 to provide a concentration of approximately $1x10^5$ spores per ml. An aliquot of 20µl of the adjusted spore suspension was added to the MEA above each of the previously marked circles.

Incubation period: All dishes were incubated at 23ºC in the dark for 24 hours between innoculation of spores and assessment.

Assessments: After 24 hours, a drop of lactophenol methylene blue was added to the agar above each circle to inhibit further germination of spores. Approximately 100 spores per circle were then examined and the incidence of germination recorded.

Analysis of data: The percentage of spores that had germinated in each circle was calculated and the mean of the three values from each replicate was used in the analysis of the data. The data were analysed by analysis of variance (following angular transformation) and differences compared using LSD.

Results and discussion:

The mean percentage of *B. bassiana* spores that germinated after storage intervals of up to 7 days following application of five fungicides and an untreated control are shown in Table 6.

In the untreated control, there was no significant difference in the percentage of spores that germinated on the four assessment dates; the overall mean being 98.2%. The results for Nimrod and Fungaflor were similar to the untreated control over the 7 day period. Scala was also similar to the untreated control but results were only available for 3 days

post-treatment. These results suggest that it should be possible to integrate all of these fungicides with *B. bassiana* in an IPM programme.

Repulse prevented germination of *B. bassiana* spores for at least 7 days. The effect of Amistar was intermediate; the percentage germination being approximately half that of the untreated controls seven days after the fungicide was applied ($P<0.05$). These results suggest that neither Repulse nor Amistar could easily be integrated with *B. bassiana* in an IPM programme.

The results for the untreated control and Repulse were consistent to those obtained in a previous experiment, which tested the immediate effect of various rates of seven fungicides (Jacobson, 2000a). However, the percentage of spores that germinated on the same day as Nimrod, Fungaflor or Scala were applied, was greater in this experiment than had been reported previously. This may have been due to a slight variation in the experimental method. In this experiment the fungicides were sprayed onto the surface of the agar, while in the previous experiment they had been incorporated into the growing medium. It is important to remember that both methods present an artificial scenario and only provide a guide to the likely compatibility of the fungicides and the entomopathogenic fungus.

Table 6. Mean percentage (angular transformed) of *B. bassiana* spores that germinated after storage intervals of up to 7 days following application of five fungicides and an untreated control.

Product	Mean percentage (angular transformed) of spores germinated on the following number of days after application of fungicides:			
	0	1	3	7
Untreated	96.7 (79.6)	99.3 (85.3)	98.4 (83.8)	98.6 (83.7)
Nimrod	97.8 (81.8)	99.7 (87.0)	99.7 (87.2)	99.4 (86.4)
Scala	97.3 (80.7)	99.2 (85.0)	99.2 (85.0)	-
Fungaflor	96.7 (79.7)	98.4 (83.2)	98.1 (82.4)	99.0 (84.5)
Repulse *	0	0	0	0
Amistar	54.4 (47.4)	83.3 (66.0)	68.4 (55.6)	53.4 (46.1)
LSD (38 d.f.) to compare angular transformed means = 3.9 ($P<0.05$). * Not included in analysis because all records were zero. - Lost due to contamination.				

PART 6 – TO IMPROVE THE KNOWLEDGE OF NATURAL HABITATS AND ACIVITY PATTERNS OF *LYGUS RUGULIPENNIS* AND *LIOCORIS TRIPUSTULATUS*

This section of the report summarises the work completed by Ms Fiona Hunter during her PhD studies in the Department of Agricultural and Environmental Science, University of Newcastle. Further information about the experimental work and more detailed results may be obtained direct from Ms Hunter.

OVERWINTERING SITES

Field

Overwintering studies were carried out on a clover plot at Close House field station (University of Newcastle (NZ 1265)) (see Jacobson 2000 for description of site). Previous studies (Jacobson 2000) indicated that adult *L. rugulipennis* may be overwintering in the vegetation in which they are found late in the season. An experiment was set up to monitor the movement of adult capsids around the clover plot at the beginning of the season.

Materials and methods:

Sticky traps

Directional sticky traps were used to determine the predominant direction of movement of capsids in the vicinity of the clover plot. Twenty sticky traps (clear plastic cylinders, 30cm high and 15 cm diameter, fixed 1.5 metres above the ground, painted with Tangle-trap© insect trap) were placed at five metre intervals around the perimeter of the clover plot. All traps were divided into sectors (north, south, east and west). Traps were checked every fortnight and the number and sex of *L. rugulipennis* individuals in each sector noted.

Tullgren funnel

Twelve samples of vegetation and topsoil (30 cm x 30 cm) were taken from a 25m by 11m section of the clover plot (see above). Insects were expelled using a Tullgren funnel.

Results and Discussion:

There was no difference between the number of individuals of either sex caught on the different sectors of the sticky traps. No *Lygus* were found in the Tullgren samples. These results suggest that the capsids were not overwintering in the clover plot.

Glasshouse

A number of commercial glasshouse sites were inspected for the presence of *Lygus* in debris such as crates and plastic surrounding the glasshouses. No *Lygus* were found on any sampling occasion at these sites.

SPRING MIGRATION

Materials and methods:

In order to assess the timing of movement of capsids into the crops a small-scale model system was set up at Close House field station. Two glasshouses (2.5m x 5.5m) were planted with salad crops (one cucumber crop, one tomato crop). Twenty plants were used in each glasshouse at the beginning of March, and replanted as necessary to maintain the quality of the crop. Each glasshouse contained 8 15cm x 10.5cm clear sticky traps coated in Tanglefoot©, which were checked weekly.

Results and Discussion:

No *Lygus* were found in either of the glasshouses throughout the duration of the experiment.

POPULATION DYNAMICS

Materials and methods:

Sampling methods are described in Jacobson (2000).

Examples of *L. tripustulatus* and *L. rugulipennis* bionomics in the 2000 field season are shown in Figures 4 and 5.

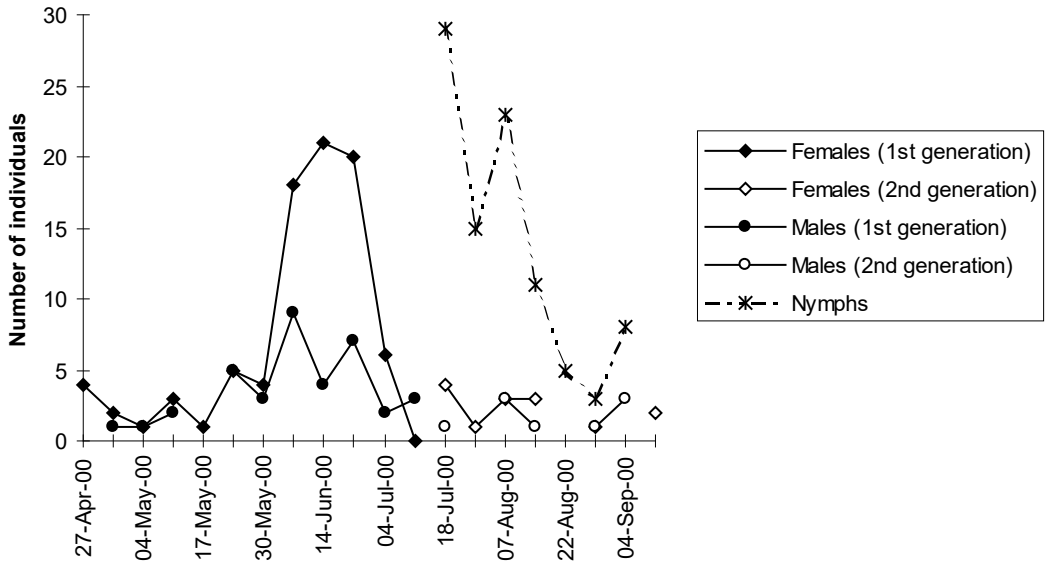


Figure 4. Example of *Liocoris* bionomics throughout 2000 field season. Mixed verge at Close House field station, Newcastle.

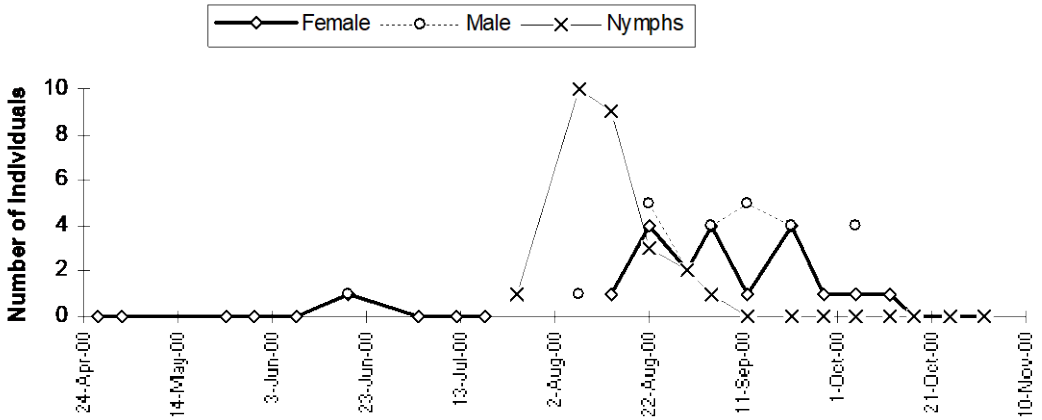


Figure 5. Example of *Lygus* bionomics throughout 2000 field season. Clover plot at Close House field station.

HOST PLANT CHOICE

Understanding why capsid bugs choose to feed on certain plants (including salad crops) and not others, and how they make that choice, are important factors in the development of successful IPM. Laboratory experiments carried out in 1999/2000 (Jacobson, 2000) were extended to examine the behavioural responses to a choice of salad or wild host leaves in a darkened choice chamber. The responses of individuals to whole plants offered in artificial light were also examined. Both *Lygus rugulipennis* and *Liocoris tripustulatus* were used in these trials. Laboratory behavioural work was then expanded to the field in an experiment using *L. rugulipennis*.

Laboratory host choice: olfaction

Materials and methods:

Insects

i) Lygus rugulipennis

Cultures of *L. rugulipennis* were maintained in the laboratory at 21°C (± 2°C), and 16:8 hours light: dark. Adults and nymphs were reared on sprouting potatoes with additions of green or runner beans *ad lib*. During the field season cultures were supplemented on a regular basis with wild caught individuals. Prior to each trial, individual 5th instar nymphs were isolated from cultures, kept individually in Petri dishes, and maintained at 21°C (± 2°C) until adult emergence. Individuals were provided with moist cotton wool, a piece of tissue paper, and a fresh green bean each day. When the adult emerged, food was withdrawn for 24 - 48 hours prior to preference tests. All adults were used in tests within 72 hours of emergence.

ii) Liocoris tripustulatus

Adult *L. tripustulatus* were collected from Close House Field. Individuals were kept separately in Petri dishes with a piece of white tissue paper and cotton wool soaked in

water. They were starved for 24 - 48 hours before the experiment. All adults were used in experiments within 72 hours of collection.

Plants

Sweet pepper (*C. annuum* var. Worldbeater), cucumber (*C. sativa* var. *sativa* Improved Telegraph) and nettle (*Urtica dioica* L., Urticaceae) plants were grown in pots in a glasshouse at Close House field station. All plants were given 20:10:10 NPK fertiliser treatments to prevent yellowing of leaves in early development. Peppers used in the enhanced nitrogen experiment were also given an application of 33% nitrogen four days before the experiment.

Determination of nitrogen levels in experimental plants

Leaves, taken from experimental plants, were freeze dried and ground to a powder for analysis of their nitrogen content using Automatic Nitrogen Carbon Analysis (ANCA). This technique measures the total nitrogen content of the sample material.

Experimental set up

Individual adults were introduced to a rectangular choice chamber (225 mm x 120 mm x 85 mm) which was lined with paper towel (replaced after each trial). Single leaves of the test species were placed at either end of the chamber. Adults were introduced to a central dish (40 mm diameter, 50 mm height) and the area containing the choice chamber and video equipment was darkened with a blackout. The majority of insects are not thought to be able to detect wavelengths above around 650 nm (red) (Hardie *et al.*, 2001), although there are exceptions (Schmitz *et al.*, 1997). Activity was recorded using a video camera (Baxall CD9242/IR), sensitive to low light levels and infra-red light from an array of light emitting diodes, and a time-lapse video recorder (Panasonic AG-6040). Capsids of both species were offered leaves of their salad and wild hosts in the following combinations:

	Wild host	**Salad host**	**Salad non-host**
L. rugulipennis	Nettle	Cucumber	x
L. rugulipennis	Nettle	x	Sweet pepper
L. tripustulatus	Nettle	Sweet pepper	x

Results and Discussion:

A number of behavioural parameters were used to assess preference, including the total time spent in contact with each leaf during a one hour period and the number of individual contacts made with each leaf. The key results are reported below. Full details of all parameters measured may be obtained from Ms Hunter.

Lygus rugulipennis

Female *L. rugulipennis* spend a greater cumulative time on cucumber leaves than nettle leaves ($W = 428.0$, $P < 0.001$), however males do not discriminate (Figure 6). Comparison of the number of separate contacts made on each leaf during a trial showed that when female *L. rugulipennis* chose cucumber first in a trial they were more likely to stay on the leaf and not make further exploratory movements around the chamber than if they chose nettle first ($W = 324.0$, $p < 0.001$). Again males did not discriminate.

Females chose cucumber leaves first in more trials than nettle leaves (Table 7), and this choice was made from a distance. Olfaction must have been used to make the choice.

Sex	Number of first contacts		χ^2 result
Female	Cucumber -23	Nettle - 7	$\chi^2 = 7.5$, $p<0.01$, n = 30
Male	Cucumber - 17	Nettle - 13	$\chi^2 = 0.3$, ns, n = 30

Table 7. *Lygus rugulipennis*: Cucumber (salad host) versus nettle (wild host). Number of times each species was chosen first in a trial.

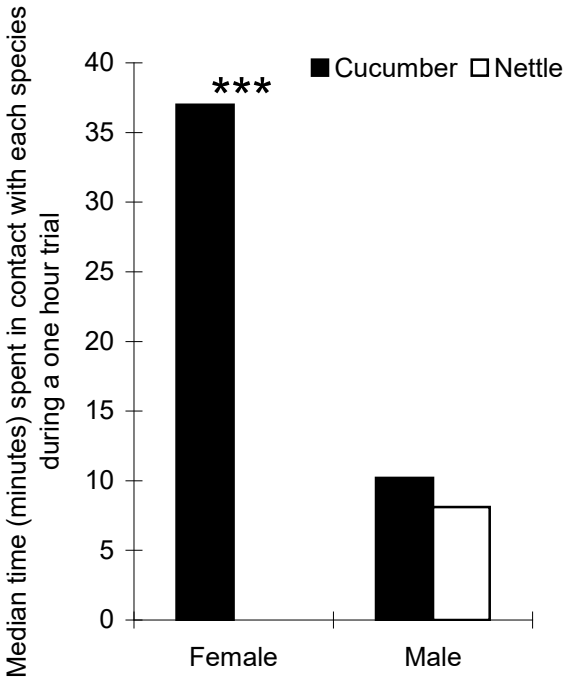


Figure 6. Median time (minutes) spent in contact with each species during a one hour trial. *Lygus rugulipennis:* Cucumber (salad host) versus nettle (wild host). Asterisk denotes significantly different results (* = P<0.001), females n = 20, males n = 20.**

<u>*Liocoris tripustulatus*</u>

Female *L. tripustulatus* spent a greater cumulative time on nettle leaves than on sweet pepper (Figure 7), however there was no difference in the number of separate contacts on either species. There was also no difference in the number of times each species was chosen as a first contact in a trial. These results suggest that *L. tripustulatus* is not exhibiting a preference based on remote olfaction, or that olfaction is only effective in combination with visual cues.

Nitrogen analysis

In both trials, the salad hosts contained more total nitrogen than the wild hosts (Figure 8). Adult *Lygus* were choosing plants with the highest nitrogen content, which may be an important factor in their host choice.

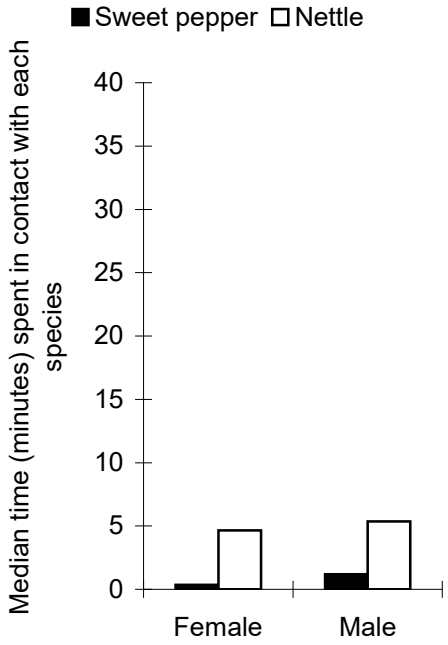


Figure 7. Median time (minutes) spent in contact with each species during a one hour trial. *Liocoris tripustulatus: Sweet pepper* (salad host) versus nettle (wild host). Asterisk denotes significantly different results (* = P<0.05), females n = 30, males n = 30.

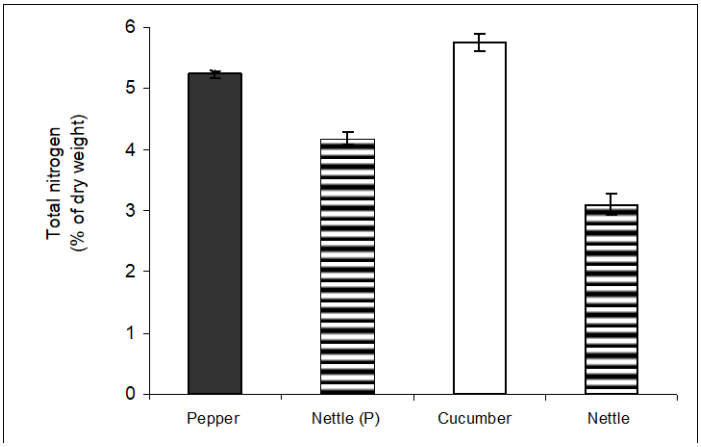


Figure 8. Comparison of nitrogen content of experimental plants used in olfactory choice tests. Error bars represent 1 S.E. of the mean (n = 3).

Laboratory host choice: plant

Materials and methods:

Experimental set up

A choice chamber (41 cm x 30 cm x 21 cm) was set up in an incubator at 21°C ± 2°C. The lower half of the chamber was covered with fleece to create a false floor on which the capsids could walk. Size matched plant cuttings were placed in the chamber so that the stem was pushed through the fleece into a water container, which kept the preparation fresh for the duration of the experiments. Individual capsids were placed in the middle of the chamber and left for one hour under artificial light to make a choice. After one hour the position of the capsid was noted.

Results and Discussion:

L. rugulipennis females and males showed no preference between cucumber and nettle when presented with plants in a lighted arena (Table 8). *L. tripustulatus* females and males both exhibited a preference for nettle over sweet pepper (Table 9).

	No. on cucumber	No. on nettle	χ^2
Lygus female	11	9	ns
Lygus male	11	9	ns

Table 8. Number of individuals on each species after one hour. Whole plant trials: *L. rugulipennis*.

	No. on cucumber	No. on nettle	χ^2
Liocoris female	5	15	$P < 0.05$
Liocoris male	5	15	$P < 0.05$

Table 9. Number of individuals on each species after one hour. Whole plant trials: *L. tripustulatus*.

Host plant choice: field experiment

Laboratory experiments showed that, using remote olfaction, *L. rugulipennis* females chose leaves with higher nitrogen content. This may be linked with the need for nitrogen for development of eggs or choosing suitable oviposition sites. A field trial was set up to test the hypothesis that female *L. rugulipennis* would occur on plants with the highest nitrogen content at certain points, or throughout the season.

Materials and methods:

Twelve plots (4 m x 4m) were sown on 3 May 2000, six with chickweed (*Stellaria media* (L.) Vill.) and six with wheat (*Triticum aestivum*) at Close House Field Station (University of Newcastle (NZ 1265). The plots were arranged in a stratified randomised block design. Three randomly selected plots of each species were given additional nitrogen fertiliser (20:10:10 N:P:K) on 24 May 2000 and 8 June 2000.

Insect samples were taken at approximately fortnightly intervals from 12 July 2000 to 17 October 2000 using a modified blower-vac. Three circular samples of 35cm diameter were taken from each plot and hand sorted in the field to collect any *L. rugulipennis* individuals. All *Lygus* spp. collected were killed and preserved in 70% ethanol. The sex of *L. rugulipennis* adults, and instar of nymphs was determined in the laboratory.

On each sampling date, three samples of plant material were collected from each plot. Only the structures on which *L. rugulipennis* are known to feed were collected, *i.e.* ears of wheat and the uppermost leaves and flowers of chickweed (Pers. Obs., Varis 1972). Plant samples were then freeze dried and analysed for total nitrogen and carbon content using Automatic Carbon Nitrogen Analysis.

Results and Discussion:

Plant nitrogen content and *L. rugulipennis* numbers in each plot over the season were analysed using the "repeated measures ANOVA" statistical test.

Although there were differences in the nitrogen content of the plots, when these changes were examined over the whole season the change could not be attributed to an interaction between species and fertiliser. Therefore the lack of detectable difference may have influenced the distribution of the capsids, which did not differ over different treatments with time.

Nymphs were only found on the chickweed plots, although there was no difference in their distribution depending on fertiliser treatment (Repeated measures ANOVA: week, $P< 0.001$; nitrogen level, ns). *Lygus rugulipennis* nymphs are unlikely to have moved between plots and their distribution can therefore be attributed to the choice of oviposition site by adult females early in the season.

The full results may be obtained from Ms Hunter.

TECHNOLOGY TRANSFER

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