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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.

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GROWER SUMMARY

Headline

• Models have been developed for predicting the timing of development of European tarnished plant bug (*Lygus rugulipennis*), strawberry blossom weevil (*Anthonomus rubi*) and strawberry tarsonemid mite (*Phytonemus pallidus* ssp. *Fragariae*)

Background and expected deliverables

Strawberries are very susceptible to many pests and diseases, many of which cannot currently be effectively controlled by non-pesticidal means. These include *Botrytis*, powdery mildew, black spot, European tarnished plant bug, strawberry blossom weevil, western flower thrips, aphids and tarsonemid mite. Correct timing/targeting of control strategies, and decisions on whether intervention is needed based on interpretation of pest monitoring or pest thresholds, depend on our understanding of pest development in relation to climatic conditions. Some developmental stages of pests may be more susceptible to insecticides than others; information on when the most susceptible stages are present would enable more effective pesticide targeting. For pests in general (unlike diseases), the developmental rate is mostly related to temperature; mathematical models are used to describe such temperature-developmental-rate relationships. These relationships will be different for different insect and mite species.

Diapausing adults of European tarnished plant bugs (capsids) overwinter on weeds or crop debris. Overwinter mortality is high, so low numbers of individuals are normally present early in the year. The first generation of the pest produced by the overwintered adults develops on weeds, and adults from this generation disperse into strawberry where a second (and possibly a third) generation occurs. The dispersal into strawberry has generally been at the time of flowering of everbearer strawberries. Capsids feeding on developing fruits cause the typical 'cat face' damage seen on everbearer strawberries. Recent observations, however, suggest that capsids may disperse to and cause damage to many other crops, including June-bearerer strawberry, raspberry and blackberry, at much earlier times than previously reported, possibly because of warm winters and springs. An understanding of the rates of development of capsids in weeds in early spring would enable growers to predict when the dispersal of first generation adults is likely to occur, and would aid in timing of placement of pheromone traps and in decisions on pesticide application timing.

Forecasting models for *Botrytis* and powdery mildew have been developed and implemented as a computer programme (in HortLink project HL0191). The project focused on the development of a holistic Integrated Pest and Disease Management system for production of strawberries which does not rely on intensive use of fungicides and insecticides during flowering and fruit development. The research work on pests in this HortLink project focused on developing alternative non-pesticidal control methods. The use of forecasting models for pests would increase the understanding of when they are likely to arrive in crops and how quickly they will develop when there. It may also be possible to use this information to develop treatment thresholds for the pest. In a current HortLink project HL01107, a model for predicting western flower thrips development is already being developed.

In this project (SF 114) the aim was to develop phenological models for capsids, strawberry blossom weevil and tarsonemid mite on strawberry and to validate the capsid model with field-collected data.

Summary of the project and main conclusions

We have developed prediction models for three strawberry pests (capsid, strawberry blossom weevil and tarsonemid mite) and incorporated them into a computer programme that already contains models for strawberry grey mould, powdery mildew and western flower thrips. New data were obtained in laboratory experiments by studying capsid development under fluctuating low temperatures to incorporate into the capsid model. To evaluate the capsid model, capsid numbers were monitored in 2010 and 2011 under both open-field and protected conditions, on weeds and strawberry plants. Predicted capsid population patterns from the model agreed well with the observed data. Thus, the capsid model can be used in practice to assist growers in managing this pest. Running the model will enable growers to identify the timing of first generation capsid adult dispersal into strawberry, or other crops, from weeds. This will aid in timing the deployment of pheromone traps to monitor populations in the crop and make it possible to target applications of insecticides against the pest. Validation of the models developed for strawberry blossom weevil and tarsonemid mite was not part of this current project; field validation will be needed before these models can be used by growers.

Financial benefits

Lygus rugulipennis (European tarnished plant bug) is a serious pest on everbearer strawberries causing crop losses by feeding on developing fruits which become deformed and unmarketable. Over 50% of fruit may be downgraded as a result of capsid feeding in unsprayed crops. The predictions made by the capsid model developed in this project

agreed well with observed catches in pheromone traps and in sweep and tap sampling on plants throughout the growing season. Thus the use of the model will enable growers to make decisions on whether intervention is needed based on interpretation of pest monitoring and pheromone trap catches, and to apply control strategies to accurately target this pest. In addition, different developmental stages of capsids may be more susceptible to some insecticides than others; information from the model on when these susceptible stages will be present will improve control of the pest. Thus the use of the capsid model should enable growers to make savings in insecticide applications against this damaging pest.

Action points for growers

- Growers should initially run the capsid forecasting model (and models for other pests and diseases) to gain an understanding of their use without using the forecasts for making decisions on management strategies.
- Once they have gained sufficient confidence in the use of the model, they may use the forecasts to inform their decision making.

SCIENCE SECTION

Introduction

Strawberries are very susceptible to many pests and diseases, most of which cannot currently be effectively controlled by non-pesticidal means. These include *botrytis*, mildew, blackspot, European tarnished plant bug (*Lygus rugulipennis*), strawberry blossom weevil (*Anthonomous rubi*), and strawberry tarsonemid mite (*Phytonemus pallidus* ssp. *Fragariae*). Correct timing/targeting of control strategies and decisions on whether intervention is needed, based on interpretation of pest monitoring or pest thresholds, depend on our understanding of pest development in relation to climatic conditions. Some developmental stages of pests may be more susceptible to insecticides than others; information on when the most susceptible stages are present would enable more effective pesticide targeting. For pests in general (unlike diseases), the developmental rate is mostly related to temperature; mathematical models are used to describe such temperature-developmental-rate relationships. These relationships will be different for different insect and mite species.

HDC project SF 94 / HortLink project HL0191 (completed March 2013) focused on the development of a holistic integrated pest and disease management system for the production of strawberries which does not rely on intensive use of fungicides and insecticides during flowering and fruit development. In this HortLink project, simple forecasting models for *Botrytis* and powdery mildew were developed and implemented as a computer programme; the research work on pests in this project focused on developing alternative non-pesticidal control methods. The use of forecasting models for pests would increase the understanding of when they are likely to arrive in crops and how quickly they will develop when there. It may be possible to use this information to develop treatment thresholds for the pest. One simple-yet-useful model is that based on degree-days. These models often need a base temperature (*i.e.* the minimum temperature required for development) from which to accumulate degree days. Sometimes a maximum temperature that the insect can survive).

Operating temperature-based models (e.g. degree-days) is relatively cheap and straightforward since it only needs temperature as an input, which can be provided by cheap and easy-to-use data loggers. A further advantage is that temperature can be forecast relatively accurately for 24-48 h and such forecasts can be incorporated into pest prediction. In another HortLink project HL01107 (SF 120), we are also developing a model that predicts

population development of western flower thrips (WFT); this model has also been incorporated with the disease models.

In the present project, the overall aim was to develop models for other key strawberry pests, but to focus specifically on the development and validation of the model for European tarnished plant bug (capsids). In addition to collecting data on capsid development in relation to temperatures for developing and validating the model, we also need to understand the overwintering behaviour of this pest. Diapausing adults of European tarnished plant bugs overwinter on weeds or crop debris. Overwinter mortality is high. The first generation of the pest develops on weeds, and adults from this generation disperse into strawberry where a second (and possibly a third) generation occurs. The dispersal into strawberry has generally been at the time of flowering of everbearer strawberries. Capsids feeding on developing fruits cause the typical 'cat face' damage seen on everbearer strawberries.

Recent observations however, suggest that capsids may disperse to, and cause damage to, many other crops (including June-bearer strawberry, raspberry and blackberry) at much earlier times than previously reported, possibly because of warm winters and springs. An understanding of the rates of development of capsids in weeds in early spring would enable growers to predict when the dispersal of first generation adults is likely to occur, and would aid in timing of placement of pheromone traps and in decisions on pesticide application timing.

The overall aim of this project was to develop temperature-based models to predict development of three key pests and to conduct experiments to validate and evaluate the model developed for European tarnished plant bug (capsid: *L. rugulipennis*) for accurate prediction of pest development.

Materials and methods

Development of a prediction model for L. rugulipennis

Overview

We have developed a predictive model for *L. rugulipennis*, which simulates population development from overwintering adults onwards. As this model focuses on the capsid phenology, it does not consider natural mortality at the non-adult stages. Furthermore, for the same reason, the model does not attempt to model the reproduction rate in relation to external conditions (temperature) but rather assumes a small constant reproduction rate to

estimate the timing of key phenological stages using temperature, usually recorded at hourly intervals. This model was developed primarily on the basis of data collected recently at EMR (Easterbrook *et al.* 2003).

Overwintered adults to mature adults in spring

We have made several assumptions on the overwintering phase:

- (1) only adults can overwinter
- (2) these overwintering adults are not matured enough for sexual reproduction
- (3) the overwintered adults become sexually active only in the spring after breaking diapause
- (4) breaking diapause is only dependent on temperature and day length

The model starts from the first day of a calendar year, assuming that there is a population of overwintering adults (*i.e.* 100%). There is no published information on the thresholds of temperature and day length for ending diapause. Based on published work on other *Lygus* spp. and unpublished historical catch data at EMR, the two threshold values were set at 12°C and eight hours, respectively. Daily times of sunset and sunrise are calculated using calendar date and local latitude, and are used to determine the day length; the latitude of EMR was used to calculate sunset and sunrise times. The model further assumes that on average it takes the length of accumulated 48 hours of conditions that satisfy the two threshold values for the capsids to become ready for mating.

Oviposition period

The length of the reproductive period varied greatly according to temperature for *L. rugulipennis* (Easterbrook *et al.* 2003) but the precise relationship of the egg-laying period with temperature is unknown. There was published data on the length of oviposition at a range of temperatures (17-29°C) for a related species *L. lucorum* (Men *et al.* 2008). The length of oviposition was studied for *L. rugulipennis* at 25°C in relation to various diets (Salero *et al.* 2007), and this was similar to that of *L. lucorum* at similar temperatures. Thus, we developed a new model describing the length of oviposition in relation to temperature by combining the data from these two studies:

 $R = 0.208 - 0.01467 \cdot T + 0.000299 \cdot T^2$

where R is the rate of development during the oviposition period (day⁻¹) and *T* is temperature (°C). If $T \ge 40$ or $T \le 8$, *R* is set to 0.125, namely the female adult can only lay eggs for eight days under these conditions. This model does not consider post-oviposition adult survival

because this period is much shorter than the oviposition period (Salero *et al.* 2007) and also does not contribute to the phenological stage of the next generation.

Egg hatching

The model describing the dependence of egg hatching on temperature was derived from the data published on groundsel rather than on strawberry in the study by Easterbrook *et al.* (2003). This is because critical/economic damage to strawberry crops caused by *L. rugulipennis* is mostly due to the first-generation adults, a large proportion of which are likely to have migrated from weeds (Easterbrook 1997). Thus accurate prediction of the first generation is not only important for preventing initial crop damage but also for accurate prediction of the second generation. The model is:

$$R = -0.0449 + 0.006214 \cdot T$$

where *R* is the rate of development during the egg period (day⁻¹) and *T* is temperature (°C). If $T \ge 35$ or $T \le 8$, *R* is set to 0.0.

Nymph – immature adults

The model describing the dependence of nymph development into immature adults on temperature was derived from the data published on groundsel (Easterbrook *et al.* 2003). The model is:

$$R = -0.033 + 0.004124 \cdot T$$

where *R* is the rate of development during the nymphal period (day⁻¹) and *T* is temperature (°C). If $T \ge 35$ or $T \le 8$, *R* is set to 0.0.

Pre-oviposition period

The precise relationship of the length of pre-oviposition period with temperature is unknown for *L. rugulipennis*. There are published data on the length of pre-oviposition at a range of temperatures (17-29°C) for *L. lucorum* (Men *et al.* 2008), and at three temperatures (12.8, 15.6 and 26.7°C) for *L. hesperus* (Strong and Sheldahl 1970). The length of pre-oviposition was also studied for *L. rugulipennis* at 25°C in relation to various diets (Salero *et al.* 2007); results were similar to those of *L. lucorum* and *L. hesperus* at similar temperatures. Thus, we developed a new model describing the length of oviposition in relation to temperature by combining the data from these studies:

$R = 0.0395 - 0.0057 \cdot T$

where R is the rate of development during the pre-oviposition period (day⁻¹) and T is temperature (°C). If $T \ge 35$ or $T \le 8$, R is set to 0.0. Model simulation The Fixed Boxcar technique (Wit & Goudriaan 1978) is used to generate temporal dispersion in each developmental sub-process, hence simulating the natural variability among individuals in their development. In order to use the Fixed Boxcar technique to simulate the capsid development process for a given population, it is necessary to know the average length of time required for each sub-process (*i.e.* the rate of development) as a function of temperature, and also the coefficient of variation in the developmental time. The relationship of the rate for each sub-process was given in the previous five sections.

The required number (BN) of fixed boxcars was calculated using the following equation (Wit & Goudriaan 1978):

$$BN = \left(\frac{1}{CV}\right)^2$$

where CV is the coefficient of variation in the developmental time for a specific sub-process under consideration. Based on published studies (Easterbrook *et al.* 2003; Salero *et al.* 2007) or unpublished studies at EMR at specific temperatures, CV ranged from ca. 0.1 to 0.2 depending on the developmental stage and/or temperature concerned. Thus, the estimated BN ranged from 25 to 100. Various BN values were evaluated to determine their effect on the predicted phenology; the overall effect on predicted phenology is generally small so in order to save computing time, BN was set to 40.

Development of models for strawberry blossom weevil and tarsonemid mite

We have also developed forecasting models for strawberry blossom weevil and tarsonemid mite. Both models were developed from a single published study from EMR (Easterbrook *et al.* 2003) and remain to be validated against field pest development; validation of these models was not part of this current project. Thus, further funding is needed to collect field data and validate, and, if necessary, improve these two models.

Steps to run the model

This capsid prediction model has been incorporated into the computer package developed to forecast strawberry diseases (grey mould and powdery mildew) and WFT (western flower thrips) (Figure 1).

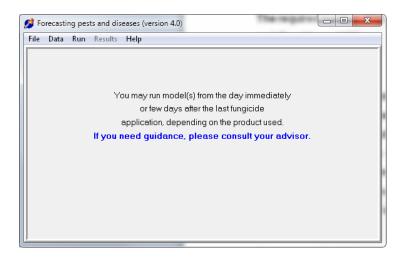


Figure 1. A screen shot of the main window of the strawberry prediction system

All the models in the system use weather data stored in the ASCII text format to generate forecasts. The ASCII format is used because all commercial weather data loggers should be able to produce data files in this format. The programme provides a very flexible data format definition facility to define the exact data format for each specific data logger (Figure 2).

💋 Weather data format	
Date time Date and time in same column? Column Date DD/MM/YY 1 1 Time H:TT 2 1	Wetness Yes? Character? Wetness threshold Column
Others 23 ◆ Temperature 23 ◆ ✓ Relative humidity? 25 ◆ ☐ Rainfall? 0 ◆	General Number of heading lines 2 Format name Davis_1 Column separator (not including space, tab or comma)
1 of €1 Previous [1] Previous	13 IsertDeleteClose

Figure 2. A screen capture of a screen form

Users may run one or more pest models (WFT, capsid, blossom weevil, and tarsonemid mite) simultaneously through a screen form (Figure 3).

Run pest models
Start Date 01/01/2013
Select a site Mansfield 🗸
Data format ADAS format
Data filename
Y:\Team Leader\Latest model\Latest bol Browse
Select model to run
Capsid Blossom weevil
Western flower thrips Tarsonemid mite
Cancel VK

Figure 3. The 'Run the model' form

After running the model(s), the model predictions can be displayed (Figure 4). The displayed predictions for capsids were for tunnel crops (covered in early April 2011) at EMR. It can be seen that there were two generations of adults [Jun-July, August-September]; note – the first blue peak shows the time when overwintered adults became active. It is possible to save the graph for filing or printing purposes by clicking the 'Save' button.

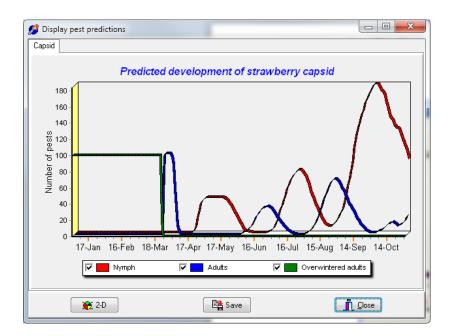


Figure 4. A screen shot of the model predictions for capsids

Calculating degree-days or degree hours

Sometimes, it is very useful to accumulate degree-days from a certain date, which can be used to predict plant or pest development. For this purpose, we have also implemented a flexible computer algorithm for calculating degree-days (or degree-hours) (Figure 5).

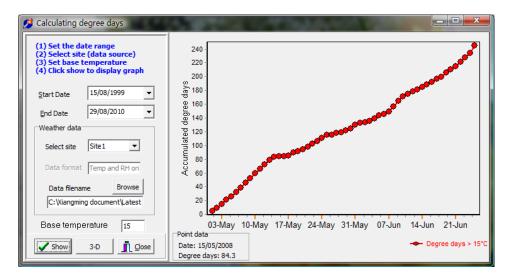


Figure 5. The 'Calculate degree-days' form.

It is also possible to display the accumulated degree-day values up to a certain date. This is achieved by single clicking a point on the line; a box titled ' Point data', containing the exact date and degree-day value, will then be shown in the bottom left (Figure 5). It is possible to start to accumulate degree-days over a new base temperature from a new date while not changing the current display. This is useful when it is necessary to calculate degree-days for multi-purposes.

To do so, double click a point on the graph from which the re-calculation of degree-days is required. A simple input window is then presented, asking for a new base temperature. The user should then enter an appropriate value and click OK; a new line will then be displayed in the graph (Figure 6). This degree-day tool enables users to define any new models based on degree days and can be a powerful tool to test new models very quickly, which may be particularly useful for consultants.

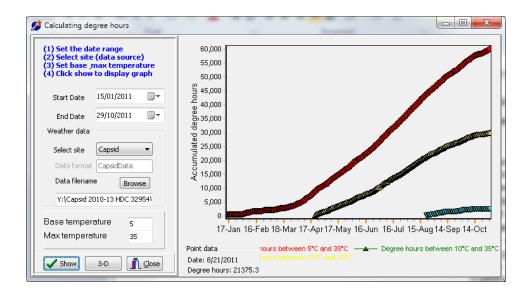


Figure 6. The 'Calculate degree-days' form showing degree-days for three different stages

Obtaining new data for the model: laboratory development studies under low fluctuating temperature

A series of experiments were conducted to study capsid development under low fluctuating temperatures (10-16°C). In the published studies, capsid development under 10°C (constant) was close to zero on groundsel and was zero on strawberry (Easterbrook *et al.* 2003). Thus experiments were needed to confirm whether this is true, since forecasting errors tend to be greatest under these turning points (extreme low or high temperatures, or around the optimum temperature).

Experiment 1: Adult female *L. rugulipennis* were introduced to a cage containing groundsel plants (*Senecio vulgaris*) and left to lay eggs. The plants were then monitored daily for the presence of recently hatched nymphs by tapping them over a white tray. One-day-old nymphs were used in the experiments. These nymphs were held individually in small Perspex boxes (approximately 16 x 10 x 8 cm). Each box contained a shoot of groundsel taken from glasshouse reared plants in a polypot of water, with the stem going into the water through a hole in the lid. Any gaps between the stem and the pot lid were plugged using absorbent towelling to prevent the nymph from drowning. Nymphs were placed in an incubator at 15°C day temperature and 10°C night temperature with a 14L:10D (light:dark; day:night) photoperiod. Nymphs were inspected and measured regularly to determine the beginning of each instar (developmental stage). Plants were also inspected for the presence of exuviae (moulted exoskeleton) to confirm the duration of each instar.

Experiment 2: The development of *L. rugulipennis* nymphs was monitored until adulthood under four temperature regimes. Adult females were allowed to lay eggs in strawberry plants (Evie-2 or Flamenco) or cut green beans in a culture cage with a 14L:10D regime at 20°C. Nymphs from these cultures were collected and used in the rearing experiments; nymphs were also collected from the field. The instar of the nymphs at collection was recorded. The nymphs were then held individually in small Perspex boxes as in Experiment 1. Each box contained two pieces of fine green bean that were cut into 4 cm lengths and then cut in half length-ways. Nymphs were placed at one of four temperatures regimes, each having a 14L:10D photoperiod regime: 16°C day temperature and 10°C night temperature; 16°C day temperature; 14°C day temperature and 10°C night temperature; 14°C day temperature regime. Nymphs were inspected daily to determine the duration of each instar.

<u>Experiment 3</u>: This experiment was set up as described in Experiment 2 except that oneday-old nymphs were used and the experimental plants were either groundsel or strawberry. A shoot of groundsel or an open strawberry flower were held in a polypot of water, with the stem going into the water through a hole in the lid as described in Experiment 1. Nymphs were reared at the same conditions as in Experiment 2, with four fluctuating temperature regimes. The duration of each instar on each plant type was recorded as above. Plant material was replaced as necessary.

<u>Experiment 4</u>: As time for development of *L. rugulipennis* eggs is unlikely to be dependent on the plant material they are laid in, adult females were placed in a cage containing fresh French bean pods as an oviposition site, and left to lay eggs at 20°C. After 24 hours the females were removed. Numbers of eggs inserted into the beans was recorded and the beans placed into cabinets with experimental conditions as described in Experiment 2 above. The time to hatch of eggs and number hatched was recorded in each temperature regime.

Field monitoring of capsid development at EMR

Capsid development under field conditions was monitored; these monitored field development data were then used to evaluate the accuracy of model predictions. In addition to data on pest developmental stage present, temperature at each site was also recorded at an interval of either 30 or 60 minutes.

2010 protocols

<u>Weeds</u>: Sweep samples were taken on weed plots between 30 April and 22 November. A standard sample was 20 passes over the selected vegetation with a 50 cm diameter sweep net. If the sample size was less than this the number of passes was recorded to enable comparisons over time to be made. Multiple samples were taken on each date and the site and main weed species present were recorded. The numbers of *L. rugulipennis* nymphs (of each instar) and adults caught in sweep samples were identified and recorded in the laboratory. Total numbers recorded from each sample date were calculated.

<u>Strawberry</u>: Tap samples were taken on an everbearer strawberry planting (DM 183) between June and November; the variety was Evie 2. The strawberry plants were tapped over a white circular tray and the numbers of *L. rugulipennis* nymphs (of each instar) and adults caught were recorded in the field.

2011 protocols

<u>Weeds</u>: Sweep samples were taken on weed plots between 16 March and 22 September as in 2010.

<u>Strawberry</u>: Tap samples were taken on an everbearer strawberry planting (DM 183) between April and September as in 2010; the variety was Evie 2. This strawberry planting was not tunnelled.

<u>Pheromone trap catches</u>: Pheromone traps were placed in a weed strip on 16 March and numbers of males caught in the traps were counted every week until early October.

2012 protocols

<u>Pheromone trap catches</u>: Pheromone traps were put out from 2 February to 21 March in two strawberry crops and in a grassy orchard area next to an old weed plot that had been ploughed in but may have been an over-wintering site for the capsids. The aim was to determine the early flight time for overwintered adults in order to estimate the conditions for breaking diapause.

<u>Development of overwintered females</u>: Ovarian development of adult females was assessed to obtain information on the potential timing of initial oviposition in spring. Females collected from the field were dissected under a microscope and the presence of developed eggs recorded.

Other field monitoring of capsid development

In addition to monitoring undertaken as part of this project, capsid monitoring was also carried out as part of two HortLink projects (HL0191 / SF 94 and HL0184 / PC/SF 276). These data were included in the evaluation of the model outputs.

<u>Tap sampling</u>: Samples were taken from a commercial strawberry planting of the cultivar Amesti from April to August in 2011. This planting was tunnelled as in normal commercial practise. Samples were also taken in 2012.

<u>Pheromone trap</u> catches: Traps were placed in two experimental tunnels at EMR and in commercial sites in Kent in 2010, 2011 and 2012. Numbers of males caught in the traps were counted every week.

Results

Laboratory development studies under low fluctuating temperature

Experiment 1: The mean body length of *L. rugulipennis*, from the head to the base of the abdomen (x10 magnification) at the first observation after moulting is shown in Table 1 and the duration of each stage at 15°C day temperature with 14 hours light and 10°C night temperature with 10 hours dark in Table 2. The number of insects followed through their development is shown in both tables. Although initially 16 individuals were set up there was a high mortality rate during development; only the nymphs that developed to at least instar 5 have been included in the results.

Lygus stage	Number recorded	Mean body length at moult (relative units)
Instar 2	5	1.35
Instar 3	5	1.72
Instar 4	5	2.30
Instar 5	5	3.08
Adult	3	5.17

Table 1. Relative body size of different instars of L. rugulipennis nymphs after moulting

Table 2. The mean number of days taken for development of each nymphal instar of Lygus
rugulipennis at 15°C and 10°C temperature reared in 14L:10D photoperiod on
groundsel

Instar	Number recorded	Days	
1	5	12.1	
2	5	10.6	
3	5	9.6	
4	5	11.4	
5	3	15.8	
Total number of day	s from instar 1 to adult	59.5	

Experiment 2: The mean duration of each nymphal instar (days) at different temperature regimes is shown in Table 3. Individuals were at different developmental stages when they were collected and did not all complete development before dying. In addition, mortality is high in young nymphs. Therefore numbers of individuals contributing to the mean duration are given in parentheses.

Table 3. The mean duration of each nymphal instar of Lygus rugulipennis in differenttemperature regimes with 14L:10D reared on bean. Numbers contributing to themean are shown in parentheses

	Duration (Duration (days)					
Temperature	Instar 2	Instar 3	Instar 4	Instar 5	for instars 2- adult		
16°C L:16°C D	6.6 (3)	8.5 (4)	8.1 (8)	12.6 (10)	35.8		
16°C L:10°C D	11.3 (3)	9.5 (6)	9.4 (8)	15.3 (10)	45.5		
14°C L:14°C D	13 (1)	10.3 (6)	11.6 (8)	15.7 (9)	50.6		
14°C L:10°C D	9.5 (4)	12.4 (7)	16.3 (10)	19.1 (10)	57.3		

As expected, the nymphs that were held at a constant 16°C developed more quickly than those at the other temperature regimes. Total development time from the beginning of instar 2 to adults was around 36 days at a constant 16°C compared to 46 days at 16°C:10°C; 51 at constant 14°C and 57 at 14°C:10°C.

Experiment 3: As in earlier experiments, mortality of nymphs was very high (Tables 4 and 5). Twenty-eight or 29 one-day-old nymphs were used in each temperature regime in the groundsel experiment and 9-13 survived to moult to the next instar. On strawberry 21 or 23 individuals were used in each temperature regime and only 8-10 survived to the next instar. On strawberry only one individual survived to the adult stage (Table 5), whereas 12 survived on groundsel. In the experiment on groundsel there was no apparent effect of set temperature regime on development time from egg hatch to adults (Table 4). Only one individual developed to the adult stage on strawberry (Table 5). However, when the

temperature loggers in the CE cabinets were downloaded it became apparent that the overall mean temperatures the nymphs had been subjected to were not as set in the four treatments due to the functioning of the cabinets. Mean daily temperatures for the four treatments should have been 12.3, 14.0, 13.5 and 16.0 but were actually 14.0, 15.3, 14.4 and 15.5 for the 14/10, 14/14, 16/10 and 16/16 treatments respectively. Thus it is not possible to draw any conclusions about effects of temperature on total development time of nymphs in this experiment. However, the results have given us new information on development rates at the actual temperatures obtained for inclusion in the model.

Table 4. The mean duration of each nymphal instar of Lygus rugulipennis in the set temperatureregimes with 14L:10D reared on groundsel. Numbers contributing to the meanare shown in parentheses

Temperature	Duration	Duration (days)					
	Instar 1	Instar 2	Instar 3	Instar 4	Instar 5	duration	
16°C L:16°C D	10.4 (13)	9.4 (7)	13.2 (5)	11.6 (5)	18 (2)	62.6	
16°C L:10°C D	11.4 (9)	10 (3)	9.5 (2)	13 (2)	17 (2)	60.9	
14°C L:14°C D	10.6 (10)	8 (7)	8.2 (7)	10.7 (3)	13.7 (3)	51.2	
14°C L:10°C D	11.2 (12)	10.4 (5)	10 (5)	12 (5)	16.8 (5)	60.4	

Table 5. The mean duration of each nymphal instar of Lygus rugulipennis in the set temperatureregimes with 14L:10D reared on strawberry. Numbers contributing to the meanare shown in parentheses

Temperature	Duration (day	Duration (days)					
	Instar 1	Instar 2	Instar 3	Instar 4	Instar 5	duration	
16°C L:16°C D	11.2 (9)	11.2 (5)	12.5 (4)	16 (3)	-	-	
16°C L:10°C D	9.4 (10)	10 (7)	11.2 (5)	13.5 (4)	24 (1)	68.1	
14°C L:14°C D	10.8 (8)	9.4 (5)	7.8 (4)	-	-	-	
14°C L:10°C D	12.8 (8)	8.8 (6)	9.2 (5)	14 (5)	-	-	

<u>Experiment 4</u>: The first eggs to hatch were those in the constant 16°C temperature regime (Figure 7). Although the first eggs in the 16°C:10°C regime hatched on day 23, the hatching period in this treatment was longer than in any of the others. All eggs had hatched in the two constant temperature regimes within two days of each other. Hatching was considerably delayed in the 14°C:10°C treatment. The percentage of eggs that successfully hatched was also different between treatments: at a constant 16°C all eggs hatched (out of a total of 50); at 16°C:10°C 64% hatched (out of a total of 76); at a constant 14°C 100% hatched (out of a total of 52) and in the 14°C:10°C treatment only 23% hatched (out of a total of 52).

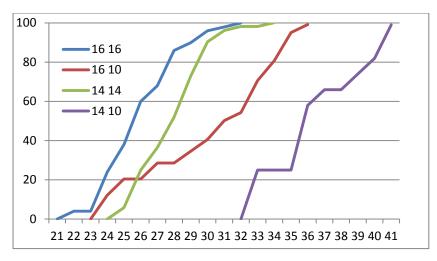


Figure 7. Cumulative percentage egg hatch in days of *L. rugulipennis* in different temperature regimes with 14L:10D

Field monitoring at EMR and model evaluation

2010 results

<u>Weeds</u>: The species sampled were mainly fathen (*Chenopodium album*), groundsel (*Senecio vulgaris*) or a mixture of cornflower (*Centaurea cyanus*), corn chamomile (*Chrysanthemum segetum*) and corn marigold (*Anthemis arvensis*); this mixture was sown for a different experiment. Total number of sweeps taken per date ranged from 29 (on 13 July) to 240 (on 30 April); most dates had 100 or more sweeps. For ease of comparison throughout the season, numbers were normalised to 20 sweeps (Figure 8 and Table 6). Only adults were caught in the first samples taken on 30 April, these being the overwintered adults. By 24 May, instar 3 nymphs were present (Table 6), indicating that eggs had probably been laid in April by the overwintered adults. Nymphs were found in low numbers in samples until 5 July; this is likely to be the end of the first generation nymphs, with a peak of adults of this first generation caught on 13 July (Figure 8). Nymphs were again found in low numbers at the end of July, with relatively higher numbers of instars 1, 2 and 3 in the samples on 3, 11 and 18 August, and relatively higher numbers of instars 4 and 5 in samples taken on 23 September, with a subsequent peak of second generation adults on 30 September (Figure 8 and Table 6). Adults were still active into November.

Date	Nymph	Adults				
Dale	1	2	3	4	5	Aduits
30 April	0	0	0	0	0	1.2
14 May	0	0	0	0	0	0
24 May	0.2	0.4	0.6	0	0	1.4
3 June	0	0	0	0	0	1.4
16 June	0	0	0.2	0.6	0.4	1
23 June	0	0.4	0.2	0	0.4	2.8
5 July	0	0	0	0	0	0.8
7 July	0	0	0	0	0	1.8
13 July	0	0	0	0	0	15.8
20 July	0	0.2	0.2	0	0.2	3.8
28 July	0.2	0.8	1.2	0.8	0.6	3.2
3 August	1.8	3.6	3	2.2	0.6	4.6
11 August	7.6	9.6	10.6	4.4	1	5.6
18 August	12	12.2	13.6	13.2	5.2	3.8
23 September	0	0.2	1.6	5.6	52.2	126
30 September	0	0	0.2	0	4.2	161
7 October	0	0	0	0.2	2.6	102
21 October	0	0	0	0	0	25.8
28 October	0	0	0	0	0	14
11 November	0	0	0	0	0	3.2
22 November	0	0	0	0	0	8.4

Table 6. Numbers of Lygus rugulipennis nymphs (of each developmental stage) and adultsrecorded in sweep samples on weed plots at EMR in 2010; samplesnormalised to 20 sweeps

<u>Strawberry</u>: Counts were normalised to 160 taps. No *L. rugulipennis* were found in tap samples on strawberries until late July. First and second instar *L. rugulipennis* nymphs were caught in July (Figure 8 and Table 7), indicating that adults had laid eggs in the crop earlier in July. Nymphs present in July and August are responsible for the damage seen to fruit in everbearer plantations. The overall pattern of development of the different life stages from strawberry was similar to that on weeds. However, there appeared to be more local variation in the capsid counts on strawberry than on weeds (Figure 8).

Date	Nymphal stage					A
	1	2	3	4	5	Adults
7 June	0	0	0	0	0	0
23 June	0	0	0	0	0	0
7 July	0	0	0	0	0	0
20 July	12	4	0	0	0	2
28 July	0	8	0	0	0	24
11 August	28	20	28	12	4	28
18 August	6	12	30	8	6	8
28 August	16	16	16	16	0	16
2 September	8	16	24	18	14	8
9 September	2	4	16	20	8	12
23 September	0	2	6	10	40	18
24 September	0	4	0	8	0	20
30 September	0	6	10	4	12	22
7 October	0	2	0	2	0	8
21 October	0	0	0	0	2	10
28 October	0	0	0	4	2	48
10 November	0	0	0	0	0	4
24 November	0	0	0	0	0	6

Table 7. Numbers of Lygus rugulipennis nymphs (of each developmental stage) and adultscaught in tap samples on a strawberry plot (DM 183) at EMR in 2010;samples normalised to 160 taps

<u>Model predictions</u>: Predicted timing for adults (excluding overwintered adults) and nymphs are shown in Figure 8, together with field collected data. The predicted times were very close to those observed for adults and nymphs on both weeds and strawberry.

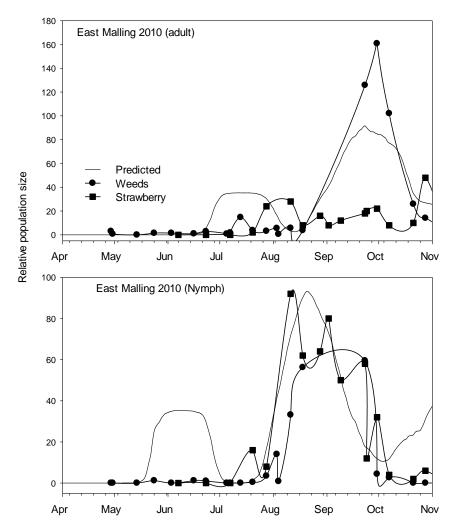


Figure 8. Predicted capsid developmental stages and the observed numbers of *Lygus rugulipennis* nymphs and adults recorded in sweep samples on weeds and tap samples in strawberry plots at EMR in 2010; sampled counts were normalised to 20 sweeps on weeds and 160 taps on strawberry

2011 results

<u>Weeds</u>: The species sampled were mainly fathen (*Chenopodium album*), groundsel (*Senecio vulgaris*) and weeds such as *Matricaria* spp. For ease of comparison throughout the season, numbers of *L. rugulipennis* caught were normalised to the equivalent of 100 sweeps. Only adults were caught in the first samples taken on 16 March (Table 8), these being the overwintered adults. Both males and females were caught early in the season. Later in the season there was an approximate 50:50 ratio of males and females. By the middle of May, instar 2 nymphs were present, indicating that eggs had been laid in April by the overwintered adults. The adults found at this time are likely to be overwintered adults. By early June mainly fifth instar nymphs and adults were caught. Second generation nymphs were found between mid-July to early-September. Relatively higher numbers of instar 1, 2 and 3 nymphs were found from 13 July to 3 August, and relatively higher numbers of instar 4

and 5 nymphs were found in samples taken on 11 August to 4 September. There was a peak of first generation adults in July and of second generation adults in mid- to late-September. These peaks can be clearly seen in Figure 9.

Date	Nymph	۸ ماریاده				
	1	2	3	4	5	Adults
16 March	0	0	0	0	0	0.5
30 March	0	0	0	0	0	0.5
05 April	0	0	0	0	0	0
12 April	0	0	0	0	0	0
20 April	0	0	0	0	0	0
28 April	0	0	0	0	0	0
03 May	0	0	0	0	0	0.6
12 May	0	0	0	0	0	0
18-19 May	0.2	0.5	0	0	0	2.3
01 June	0	0.1	0.1	0.4	1.1	3.6
08-9 June	0	0	0.2	0.3	1.2	6.4
14 June	0	0	0	0	0	1
29 June	0	0	0	0	0.2	0.8
09 July	0	0	0.3	0	0.5	1.8
13 July	0	0.3	0.2	0.2	0.1	1.5
19 July	0	0.3	0.8	0.8	0.3	2
28 July	0.8	1.5	2.8	1.8	0.8	0.3
03 August	8	6	3.5	1.5	3.5	5
11 August	0.7	1.3	2.2	5.5	10.5	2.1
01 September	0	0.3	0.8	0.5	3.2	4.6
04 September	0	0.2	0.4	0.3	1.2	5.6
14 September	0	0	0	0	1.1	18.2
22 September	0	0	0.3	0.5	2	13.5

Table 8. Numbers of Lygus rugulipennis nymphs (of each developmental stage) and adultscaught in sweep samples on weed plots at EMR in 2011. Numbers have beennormalised to 20 sweeps

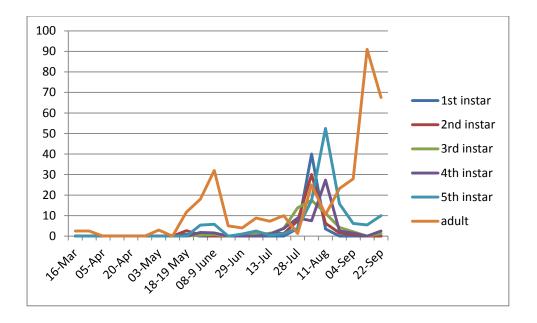


Figure 9. Development of Lygus rugulipennis populations on weeds in 2011

<u>Strawberries</u>: *L. rugulipennis* (instar 2 and 3) were found in tap samples on strawberries at EMR in the middle of May (Table 9), indicating that adults had laid eggs in the crop in late April. This is similar timing to the presence of nymphs in weeds. This strawberry plot had weeds around it and the adults had probably dispersed from these neighbouring weeds. Numbers caught were much lower in the strawberry than in the weed plots throughout the season. Slightly higher numbers of nymphs were present in July and August; the nymphs of this generation are responsible for the damage seen to fruit in everbearer plantations.

Date	Nymphal stage					
	1	2	3	4	5	Adults
20 April	0	0	0	0	0	0
3 May	0	0	0	0	0	0
12 May	0	0	0	0	0	0
19 May	0	1.6	1.6	0	0	2
8 June	0	0	0	0	0	0
14 June	0	0	1.6	0	0	1.6
29 June	0	0	0	0	0	0
9 July	1.6	0	0	0	0	1.6
13 July	0	1.6	0	0	0	0
19 July	0	1.6	3.2	0	0	1.6
28 July	1.6	1.6	0	0	0	3.2
3 August	1.6	9.6	1.6	3.2	1.6	0
11 August	4.8	4.8	12.8	9.6	14.4	11.2
25 August	0	0	0	0	8	4.8
5 September	0	0	1.6	0	1.6	4.8
14 September	0	0	0	0	0	3.2
30 September	0	0	0	0	0	11.2

Table 9. Numbers of Lygus rugulipennis nymphs (of each developmental stage) and adultscaught in tap samples on a strawberry plot (DM 183) at EMR in 2011normalised to 160 taps

<u>Model predictions</u>: In 2011, both sweep samples and pheromone trap catches of capsids were used. Since pheromone trap catches are more likely to represent the actual activity of capsids than sweep samples, for model validation the pheromone trap catches were used only for the adults, and the sweep samples only for the nymphs. Adults caught during the period of April-May were most likely overwintered adults. Thereafter the model correctly predicted the two generations in terms of both nymphs and adults (Figure 10). The predicted timing also agreed well with the sweep sample data (Figure 10 and Table 9).

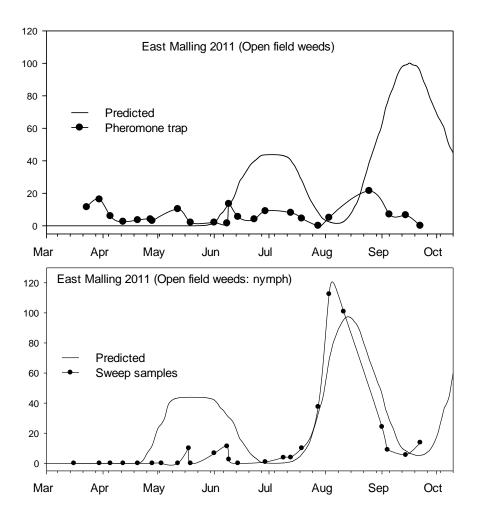


Figure 10. Predicted capsid developmental stages and the observed numbers of *Lygus rugulipennis* nymphs and adults recorded in sweep samples and pheromone trap catches on weed plots at EMR in 2011; sampled counts were normalised to 20 sweeps.

2012 results

<u>Development of overwintered females</u>: Ovarian development of adult females was assessed to obtain information on the potential timing of initial oviposition in spring. Due to the low numbers of adults found early in the season only two females were dissected to assess egg maturity. A female caught in weeds on 12 April had eight eggs which all looked well developed. A female caught two weeks before had less developed eggs, being smaller and looking 'glassy'. It is likely that the female inspected at the end of March had not fully emerged from diapause and that the female examined on 12 April had developed to the stage where egg laying could begin. In 2012 no *L. rugulipennis* males were caught in traps in February at EMR. However all three traps caught males (four-seven per trap) on 1 March, and males were also caught in the subsequent sampling weeks in March, with up to 18 males per trap.

Field monitoring at EMR and model evaluation

<u>Pheromone trap catches</u>: The first males were caught at EMR in March in 2011 (Figure 11). There were two local peaks in *L. rugulipennis* catch for males: late June and late August. The observed pattern agreed well with the predicted phenology (Figure 11)

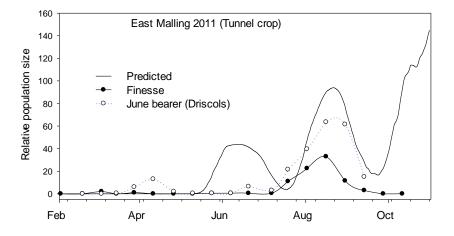
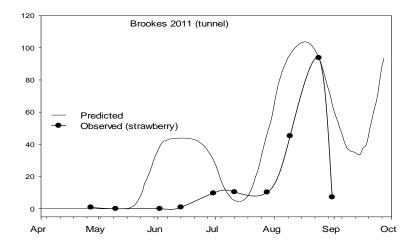
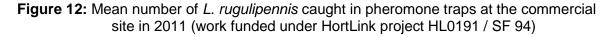


Figure 11: Mean number of *L. rugulipennis* caught in pheromone traps in two tunnel crops at EMR in 2010 (work funded under HortLink Project HL0184 / PC/SF 276

At a commercial site in Kent the first males were caught on 13 June in 2011, although with low numbers of one or two per trap (Figure 12). The peak *L. rugulipennis* catch for males was on 24 August. The observed pattern agreed well with the predicted phenology (Fig. 12).





Nymphs were not recorded on the strawberry plants until July (Table 10).

Date	Nymp	Nymphal stage					
	1	2	3	4	5	Adults	
27 April	0	0	0	0	0	0	
10 May	0	0	0	0	0	0	
2 June	0	0	0	0	0	0	
13 June	0	0	0	0	0	0	
30 June	0	0	0	0	0	0	
11 July	2	0	0	0	0	0	
28 July	0	1	6	2	2	0	
9 August	0	0	1	2	2	2	
24 August	1	0	1	0	0	0	

Table 10. Numbers of *Lygus rugulipennis* nymphs (of each developmental stage) and adults caught in tap samples of 50 plants at a commercial strawberry planting in 2011

Discussion

The present model was developed primarily based on three published studies, two on L. *rugulipennis* (Easterbrook *et al.* 2003; Salero *et al.* 2007) and one on *L. lucorum* (Men *et al.* 2008), and on unpublished data obtained at EMR. Although a web-based system for pest and disease predictions in Norway (called VIPS) contains a model for *L. rugulipennis*, its actual formulation (and indeed assumptions) is unknown. Because of this lack of information, we had to make several key assumptions related to overwintering, and minimum and maximum temperatures for development. Despite this, the model performed really well in predicting the generation times, which agreed with monitored population density data in open field or under protection in 2010 and 2011. The present model only forecasts the generation timing and not the population density. To forecast actual population density, information on several key biological aspects is needed, but is currently not available, including the relationship of fecundity and mortality with external conditions and control measures applied. This aspect of modelling capsid population growth did not form part of the current project.

The developmental rate of L. *rugulipennis* is known to be slower on strawberry than on groundsel (Easterbrook *et al.* 2003), a common weed within and/or around strawberry crops in the UK. Nevertheless, we used the capsid development data on groundsel to develop a prediction model for controlling capsid on strawberry for two reasons. First, it is generally believed that capsid adults overwinter on weeds. These overwintered adults lay eggs on weeds in spring; the resulting nymphs then feed on weeds. These adults may then migrate to strawberry and cause damage to the crop. Subsequently, they lay eggs on strawberry plants as well as other host plants. Nymphs from these eggs will then cause further damage to the crop. Thus accurate prediction of the development of the first generation of adults on

weeds is critically important for timing control measures on strawberry. Secondly, common weeds (such as groundsel) are usually present around and/within strawberry plantings. Hence predicting capsid development on groundsel over the season is as important as on strawberry.

In developing temperature-based models for predicting pest development, one of the key pieces of information needed is the threshold for development, *i.e.* minimum and maximum temperatures for development. For adults to break diapause (becoming sexually active in spring), we used a threshold of 12°C for 48 hours with day length over eight hours. This temperature threshold was based on data obtained for other Lygus species. For subsequent development stages from egg-hatching to adults, a simple linear relationship with temperature was used to estimate the developmental rate with a minimum temperature set to 8°C. This minimum temperature for development agrees well with a threshold value of 7.9°C for L. lineolaris (Ugine 2012), another strawberry pest. The development rate during the egg-hatching and nymph development periods at low temperatures predicted by the present model is also consistent with the data obtained with L. rugulipennis under fluctuating temperatures in the laboratory in the present project. However, there is no information available on the maximum temperature for L. rugulipennis development. Here, we assumed 35°C. There was no detailed information on the relationship of the length of the oviposition period with temperature for L. rugulipennis, but we used the non-linear relationship developed for L. lucorum (Men et al. 2008), which suggested that the oviposition period is shorter at both high and low temperatures. This general pattern of the oviposition period with temperature is similar to that of L. lineolaris (Ugine 2012). Further research is needed to obtain developmental data on these aspects for L. rugulipennis. Nevertheless, the fact that the model predictions agreed well with the field observations suggests that the model assumptions are not far from the true biology of L. rugulipennis.

The model predicted two clear generations, but the first generation was often not pronounced for the observed data. This is most likely due to the fact that the overwintered capsid population size is very small. For example, only 2-11% of *L. rugulipennis* adults survived winter on sugar beet in Finland (Varis 1972). In addition, control measures applied to strawberry may also affect capsid population density, which may sometimes cause difficulties in identifying peaks in capsid population size.

In summary, the present model appeared to be able to predict *L. rugulipennis* generation times accurately. Therefore, it should be able to be used in practice to assist in interpreting trap catches and decision making as regarding control measures for this pest.

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