Project title:	Outdoor lettuce: forecasting and control of Sclerotinia
Project number:	FV 294
Project leader:	Dr Caroline Young, ADAS
Report:	Annual report, 31 September 2007
Previous report	None
Key staff:	Dr Caroline Young, ADAS, Plant pathologist Dr Laura Fawcett, ADAS, modeller Dr John Clarkson, Warwick-HRI, Sclerotinia expertise and inoculum
Location of project:	Site 1: G's; J B Shropshire & Sons, Hainey Farm, Barway, Ely, Cambs CB7 STZ, field location GR TL 630 792, Lark Bank, nr Prickwillow Site 2: Merrymac Salads, Mudds Drove, Three Holes, Wisbech, Cambs PE14 9JU,
	field location GR TL 672 897, nr Feltwell
Project coordinator:	John Sedgwick, Kettle Produce, Balmalcolm, Cupar, Fife KY15 7TJ.
Date project commenced:	1 October 2006
Date project completed (or expected completion date):	31 September 2008
Key words:	Field lettuce, Sclerotinia, fungicides

Whilst reports issued under the auspices of the HDC are prepared from the best available information, neither the authors nor the HDC can accept any responsibility for inaccuracy or liability for loss, damage or injury from the application of any concept or procedure discussed.

The contents of this publication are strictly private to HDC members. No part of this publication may be presented, copied or reproduced in any form or by any means without prior written permission of the Horticultural Development Council.

The results and conclusions in this report are based on an investigation conducted over a one-year period. The conditions under which the experiments were carried out and the results have been reported in detail and with accuracy. However, because of the biological nature of the work it must be borne in mind that different circumstances and conditions could produce different results. Therefore, care must be taken with interpretation of the results, especially if they are used as the basis for commercial product recommendations.

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.

Dr Caroline Young Plant Pathologist ADAS UK Ltd	
Signature	Date
Dr John Clarkson Plant pathologist Warwick HRI	
Signature	Date
Report authorised by:	
Dr W E Parker	
Horticulture Research & Consultancy Manager	
ADAS UK Ltd	
Signature	Date
[Name]	
[Position]	
Warwick HRI	
Signature	Date

TABLE OF CONTENTS

GROWER SUMMARY	1
HEADLINE	1
BACKGROUND AND EXPECTED DELIVERABLES	1
SUMMARY OF THE PROJECT AND MAIN CONCLUSIONS	2
Development and evaluation of the Sclerotinia forecasting model	2
Field experiments to compare Sclerotinia control using different fungicides	5
FINANCIAL BENEFITS	8
ACTION POINTS FOR GROWERS	9
SCIENCE SECTION	10
INTRODUCTION	10
MATERIALS AND METHODS	14
Development of the Sclerotinia forecasting model	14
Sclerotia germination model – initial derivation	15
Sclerotia germination model – modifications for forecasting	16
Replacing soil water potential threshold with a threshold triggered by a tem	perature-
rainfall function	17
Estimating the mean time to reach 10% sclerotial germination	18
Germination Temperature Threshold	18
Different isolates of S. sclerotiorum	19
Fleeced Crops	19
Model validation	20
Model Summary and Data Requirements	20
Field site details	21
Sclerotinia sclerotiorum isolates and production of sclerotia	21
Weather data loggers for field experiments	22
Sclerotinia inoculum for field experiments	23
Field experiments to evaluate the Sclerotinia forecasting model	23
Field experiments to compare Sclerotinia control using different fungicides	24
RESULTS	27
Field experiments to evaluate the Sclerotinia forecasting model	27
Predictions of time to reach 10% sclerotial germination (T10)	27
Sclerotial germination in grids	30
Diseases in the modelling experiments	30
Field experiments to compare Sclerotinia control using different fungicides	32
Progress of lettuce growth	33

Sclerotinia disease	
Downy mildew	
Botrytis	
Lettuce yields	
DISCUSSION	
Sclerotinia forecasting model	
Sclerotinia, downy mildew and Botrytis control with diffe	erent fungicides 42
TECHNOLOGY TRANSFER	
REFERENCES	
APPENDIX 1	
Applications to commercial crops at G's and Merrymac	sites, 2007 45
Site 2: G's site, applications made to Buckinghams Fiel	ld, lettuce crop April 2007 51
APPENDIX 2	
Sclerotial germination model: replacing soil water poter	ntial threshold with a threshold
triggered by a temperature-rainfall function	

GROWER SUMMARY

Headline

- A forecasting model for Sclerotinia in outdoor lettuce was successful in the first year for predicting germination of *Sclerotinia sclerotiorum* inoculum in soil.
- In a separate fungicide experiment, no fungicides prevented downy mildew infection, but Amistar (azoxystrobin) treated lettuce had the lowest percentage leaf area affected by downy mildew. Switch (cyprodinil + fludioxonil) gave the best Botrytis control. Levels of Sclerotinia infection were too low to determine significant fungicide effects. Note: Both these products are currently approved (Nov '07) – growers should check the approval status of all products before application is made.

Background and expected deliverables

Sclerotinia disease is a common problem in outdoor lettuce, sometimes causing very high losses, and a large proportion of crops are treated with fungicide routinely. The fungus produces resting bodies in infected plants, called sclerotia. These sclerotia become incorporated into soil and can germinate the following year in spring to produce apothecia, the fungal fruiting bodies which release the spores that infect lettuce. One problem with control of Sclerotinia in outdoor lettuce is the difficulty of timing fungicide applications or justifying omission of sprays. There is good potential to improve fungicide timing using a forecasting model that predicts when sclerotia will germinate. The background data on which to base a forecasting model have been generated in a previous Defra-funded project (HH3215TFV, 'Forecasting Sclerotinia in field-grown lettuce'), in which the environmental conditions leading to germination of sclerotia were determined. In the current HDC project, the aim is to produce and field-test a model with predictive capability for sclerotial germination. At the end of this project, the Sclerotinia model will be incorporated into MORPH, the software used to operate the decision support models for various pests and diseases, developed at Warwick-HRI.

The number of fungicides for use on lettuce against Sclerotinia, and the number of applications permitted per crop, is limited. Therefore, in addition to field experiments to test the Sclerotinia forecasting model, fungicide experiments were set up to test various products for efficacy against Sclerotinia. Downy mildew and Botrytis infections were also assessed.

The objectives of this project are as follows:

- 1. Develop a Sclerotinia disease forecasting model and incorporate it into the disease forecasting system MORPH (developed at Warwick-HRI).
- 2. Evaluate the efficacy of Sclerotinia disease control on lettuce using fungicide spray applications timed according to the forecasting model.
- 3. Compare the efficacy of different fungicides and a biological control agent for Sclerotinia control.

This project is at the start of the second (final) year, in which the forecasting model will be further tested and incorporated into MORPH, and further fungicide trials will be completed.

Summary of the project and main conclusions

Development and evaluation of the Sclerotinia forecasting model

The model for sclerotial germination needed to be adapted to operate in forecasting mode to be of practical use. Also, some of the environmental data inputs needed to be replaced with simpler data where possible, e.g., soil water potential is an important factor in sclerotial germination, but requires specialised probes. Therefore a substitute factor based on a rainfall and temperature calculation was developed.

In summary, the forecasting model developed for sclerotial germination has the following characteristics:

- The model is adapted from a non-predictive model to forecast time to 10% germination of sclerotia (T10) as an indicator of the onset of a flush of apothecia.
- The model is adapted to operate with routinely collected in-field data, recorded every half-hour. The data required are: soil temperature at 5cm depth (or air temperature if soil temperature not available), and rain. Hourly or even daily data can be used if necessary.
- The forecasting model has similar predictive capability to the original simulation model.
- The model uses historical weather data in the forecast and is updated as new data becomes available.
- The model includes an option to simulate fleeced crops.
- The model has been developed using data from one of the fastest germinating Sclerotinia isolates, to indicate the earliest onset of an apothecial flush.
- The model uses weather data from the last cultivation date in the autumn and any cultivation dates thereafter which may bring sclerotia to optimum depths for germination.

The forecasting model was tested at two sites:

Site 1: Merrymac Salads, Mudds Drove, Three Holes, Wisbech, Cambs PE14 9JU, Buckinghams field (working area 12.17 ha), Ordnance Survey (OS) Grid Reference (GR) TL 672897, nr Feltwell. Lettuce cv. Edition were planted as blocks on 15 March (4-5 true leaves) and harvested 16 May. The crop was under non-woven fleece for the whole duration.

Site 2: G's; J B Shropshire & Sons, Hainey Farm, Barway, Ely, Cambs CB7 STZ, field CW63 (working area 15.46 ha), OS GR TL 630 792, Lark Bank, nr Prickwillow. Lettuce cv. Saladin (4-5 true leaves, Scotts compost Lev B2 Bulk, 4.0 cm³ blocks) were planted on 1 March 2007 and harvested 10 May. The crop was under non-woven fleece for the whole duration. The crop was irrigated with an overhead boom once on 10 March 2007, with 15mm water.

There were 5 treatments to the Sclerotinia forecasting model experiment ('modelling experiment):

- 1. Untreated
- 2. Fungicide sprays applied as timed by the Sclerotinia model
- 3. Fungicide applied at spray time 1 (early in crop development)
- 4. Fungicide applied at spray time 1 & 2 (early and mid crop)
- 5. Fungicide applied at spray time 1, 2 & 3 (early, mid and late crop)

The fungicide used in all treatments was Signum (boscalid + pyraclostrobin). The forecasting model was initially run prior to planting based on weather data recorded on-site from 21 December 2006, to give the first estimate of the time to 10% germination of sclerotia (T10). For each further week post-planting, the weather data were downloaded, and used to re-run the prediction of T10. A prediction of T10 one to two weeks ahead of the time of running the model would initiate an alert to the ADAS site manager by email or telephone, so that preparation for treatment sprays could be made. For treatments 3, 4 and 5, plots were sprayed with Signum, at weeks 2, 4 and 6.

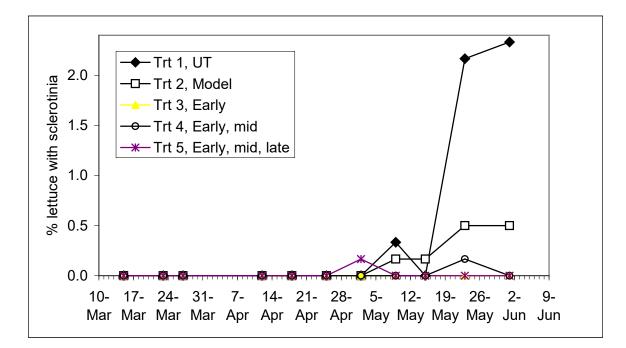


Figure 1. Merrymac 2007, modelling experiment, % lettuce with Sclerotinia disease, cv. Edition (F = 0.08). Trt 2, Model, was untreated (i.e., no spray alerts according to forecasting model); commercial harvest date was 16 May; apothecia were first observed 1 June.

In the modelling experiment, the model was successfully predicted the time of germination of sclerotia buried in the grids. There was no Sclerotinia disease seen at the G's site on any plot, but there were infected plants at Merrymac, with low incidence overall (Figure 1). No sprays for Sclerotinia at Merrymac were indicated by the forecasting model because of the dry April. The model run on 17 April predicted no germination of sclerotia until 4 May, and the crop was harvested on 16 May. The final date for prediction of apothecia at Merrymac was 18 May (Table 1). Therefore, the model was correct in suggesting no sprays were needed at Merrymac. At G's, a prediction of apothecia was made for 1 May, and consequently a spray was applied on 25 April (Table 1), but in fact, no Sclerotinia developed anywhere in the experimental plots.

Table 1. 2007, site summary for Sclerotinia modelling experiment.

	G's site	Merrymac site
Planting date	1 March	15 March
Fleece on crop (whole duration)	Yes	Yes
Harvest date (for yield)	10 May	16 May
Rain, 21 Dec '06 to 21 May '07	162	165
Irrigation	Yes	No
Date first apothecia seen in grids	7 May (max. 4%)*	1 June (max. 32%)*
Final prediction date for 10% sclerotial germination (T10)	1 May	18 May
Spray alert	Yes (sprayed 25 April)	No
Sclerotinia disease at harvest in untreated plots	0	<0.5%**

* % out of 150 sclerotia buried in grids

** Sclerotinia disease was assessed at Merrymac up to 1 June, i.e., beyond the commercial harvest date. On 1 June, untreated plots had an average of 2.3% Sclerotinia incidence.

Field experiments to compare Sclerotinia control using different fungicides

The same two field sites used in the modelling experiment described above were used for the fungicide experiment. The fungicides were applied at both sites on the same dates despite G's being planted two weeks earlier than Merrymacs, due to bad weather conditions which delayed fungicide applications at G's on the planned date. The treatments are given in Table 2. Sclerotinia, Botrytis and downy mildew were assessed, and harvest weights recorded.

Sclerotinia disease was only observed at Merrymac on cv Edition, late in the crop on 16 May (harvest) (Figure 2). Only Teldor treated plots and untreated plots had Sclerotinia disease, at low incidence (average of 0.6% or less). The incidence of Sclerotinia was too low to determine significant differences between fungicides for Sclerotinia control.

				Weeks post planting*								
No.	Product	Active ingredient	1	2	3	4	5	6	7	8	Rate	Water L/ha
1	Untreated											
2	Amistar	azoxystrobin		Х		Х		Х			1 L/ha	400
3	Signum	boscalid + pyraclostrobin		х		x		х			1.5 Kg/ha	400
4	Switch	cyprodinil + fludioxonil		х		x		х			0.8 Kg/ha	400
5	Rovral	iprodione		Х		х		х			2.3L/ha	400
6	Bayer (new) UKA383a			x		х		x			0.5L/ha	400
7	Octave	prochloraz		Х		Х		Х			200g/ha	400
8	Teldor	fenhexamid		Х		х		х			1.5 Kg/ha	600

Table 2. 2007, G's and Merrymac: fungicides, spray timing and rates for field lettuce.

*spray dates for both sites were 27 March, 12 April and 25 April.

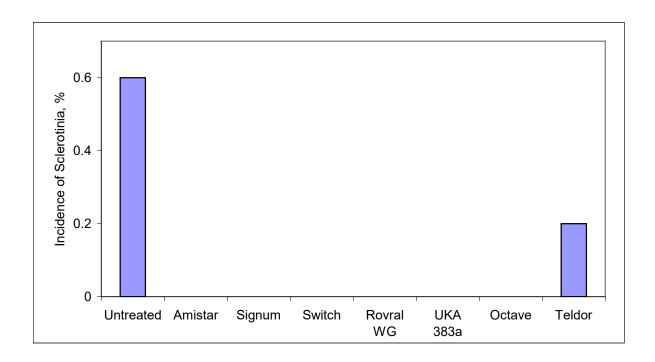


Figure 2. Merrymac 2007 fungicide experiment, % Sclerotinia disease, cv Edition, 16 May (F = 0.45).

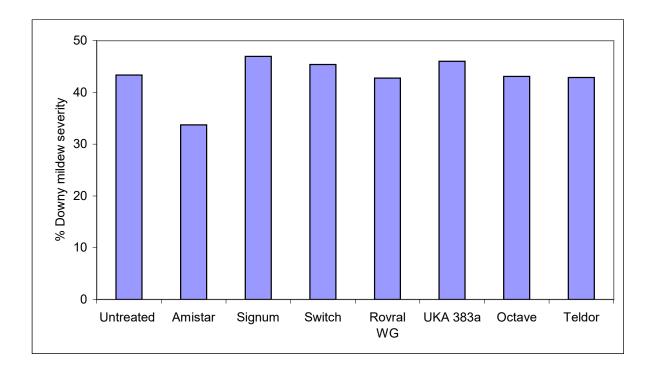


Figure 3. G's 2007 fungicide experiment, % downy mildew severity, 10 May, cv Saladin (F = 0.047).

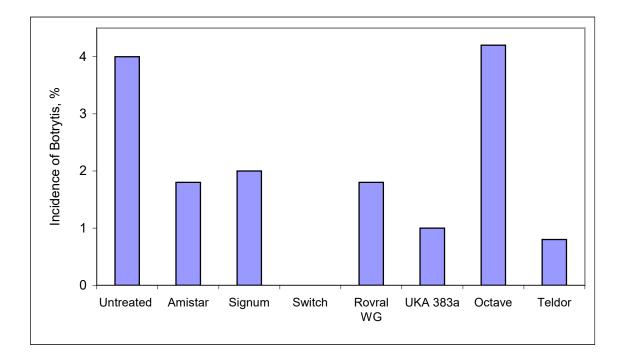


Figure 4. Merrymac 2007 fungicide experiment, % Botrytis incidence, cv Edition, 16 May (F = 0.002, SED trt = 0.98, LSD trt = 2.01).

At G's by 10 May, plants from all treatments had 35% or more leaf area affected by downy mildew, but with significant differences between treatments (Figure 3). No fungicide prevented downy mildew, but Amistar resulted in the least area affected. Downy mildew was not observed at G's on cv. Saladin.

By harvest at Merrymac, there were significant differences between fungicide treatments for Botrytis incidence, with Switch treated plants having no Botrytis, compared to 4% in untreated plots (Figure 4). There was no Botrytis recorded at G's.

Financial benefits

In summary, the cost of one fungicide spray is minimal compared to the value of the lettuce crop. Therefore, the most important financial benefit from a Sclerotinia forecasting model is to improve timing of disease control to prevent crop loss, rather than saving the costs of one or more fungicide applications.

In most years, losses are probably around 5%.

Cost of fungicide spray per hectare (ha):

Approximately £25 per ha for chemicals and £5 per ha for application costs = £30 per ha.

Value of lettuce crop per ha:

For a typical crop of 78,000 heads of lettuce per ha, worth £2.30 per dozen, which has a 62.5% cut at harvest, the value is £8,937 per ha. Therefore, a 10% loss from Sclerotinia, not uncommon, is £894. Losses of 30% or more, or even the whole crop, have been reported which has serious implications for customer supply, in addition to immediate financial losses.

On site weather data loggers are desirable, which would be an additional cost for some growers. Regional temperature data can be used, but an on-site rain gauge would be advised.

Action points for growers

- Previous weather data and dates of previous soil cultivations will be needed to run the Sclerotinia forecasting model. The weather data required are soil temperature and rainfall, from the previous autumn (September/October).
- The forecasting model should be run before lettuce planting to determine whether fungicides are needed at, or close to, planting.
- The forecasting model should be run at approximately weekly intervals, and particularly if there has been a rain or irrigation event after a period without rain.
- Cultivate entire field areas early, all at once if once if possible. This will only bring the
 sclerotia to the surface once. Any sclerotia produced in infected plants at the beginning
 of the season will not present a risk to crops until the following year (they will need to
 over-winter before they are able to germinate).
- Amistar, Signum, Switch, Rovral WG, UKA 383 and Octave all had some activity against Sclerotinia, but 2007 was unusually dry in April with low Sclerotinia risk. Therefore, the experiment did not provide a good test for efficacy against Sclerotinia.
- Amistar had the most activity against downy mildew in terms of % leaf area affected per plant. No fungicide tested prevented downy mildew infection.
- Use resistant or partly resistant cultivars for downy mildew if the disease is a known problem.
- Switch was the only fungicide to prevent Botrytis, but in 2007 the highest infection was only 4% of plants/plot affected.
- Fungicides from different chemical groups will need to be alternated, according to label instructions. This is to avoid the build-up of resistance to fungal pathogens. For example, Signum and Amistar are within the same group of fungicides (QoI fungicides*) and should not be used consecutively.

*(Qol = Quinone outside inhibitor)

SCIENCE SECTION

Introduction

Most Sclerotinia disease on field vegetables in the UK is caused by the fungus Sclerotinia sclerotiorum, which is the species referred to here as 'Sclerotinia'. Sclerotinia survives in the soil as resistant resting bodies called sclerotia, which can remain viable for years. Under the right soil conditions, usually in spring, the sclerotia brought close to the soil surface by tillage operations germinate to produce small mushroom-like apothecia. These release airborne ascospores which infect plants. The fungus then grows within infected plants and sclerotia develop which subsequently become incorporated into soil to begin the disease cycle again. Sclerotinia disease in field lettuce tends to occur each year at low to moderate incidence, but with occasional severe outbreaks with heavy crop losses. Currently it is difficult to predict when these epidemics will occur, and hence decisions about the economics of treatment are hard to make. Lettuce crops infected with Sclerotinia rot quickly, either in the field or in store. A range of other field vegetables are also susceptible to Sclerotinia, including carrots, celery, dwarf beans, runner beans, brassicas and broad beans. Arable crops, including oilseed rape, peas, potatoes and field beans also frequently become infected, and the large area of some of these crops means that they are an important potential source of Sclerotinia inoculum for field vegetables.

The incidence of Sclerotinia disease in field lettuce crops is variable, e.g. losses in early plantings of field lettuce in Cheshire can be up to 30%, while later plantings have little or no disease (Young *et al.*, 2004). In recent years Sclerotinia has been increasingly reported in field vegetables, and a high proportion of these crops are treated with fungicides. For example, in 2003, 89% (by area) of lettuce & endive, and 85% of carrots, parsnips & celery were treated (CSL Pesticide Usage Survey, 2003). Overall, fungicide use on vegetables is increasing, e.g., in 2003, in terms of area, fungicide use increased almost threefold compared with 1999. A reduction in fungicide use is desirable and would be beneficial for the environment and the economics of lettuce production.

The main problems with fungicidal control of Sclerotinia are the timing of fungicide applications to achieve good control, and the selection of an effective fungicide product. There is good potential to improve the timing of fungicide applications by developing a Sclerotinia disease forecasting model for practical use by growers. The number of fungicide applications that can be made to a lettuce crop is limited and therefore available treatments have to be used effectively. However, as *S. sclerotiorum* spores are released from

apothecia which can appear at different times each year, determination of the optimum time to spray can be a major problem for growers.

There are few published forecasting systems for Sclerotinia. A risk assessment scheme based on factors including weather, previous crop and previous disease has been produced for Sclerotinia on oilseed rape in Sweden (Twengstrom *et al*, 1998) and a system is also available for Sclerotinia in UK oilseed rape (developed by ADAS) and is available on the HGCA website (www.hgca.com/research/OSRWeb/Pages/osrindex.htm). However, these schemes are not directly applicable to leafy crops such as lettuce because Sclerotinia infects oilseed rape primarily via flower parts, which is not part of the Sclerotinia life cycle in lettuce. One commercial forecasting model targeted at simulating the behaviour of Sclerotinia on vegetable crops and in particular carrot has been produced in order to aid fungicide spray decisions, but currently it is not widely used and has not been demonstrated so far to be useful in lettuce crops.

There is therefore a need for an effective forecasting model for Sclerotinia disease for use in field lettuce. To address this, the Defra project HH3215TFV (Clarkson *et al.*, 2005) aimed at developing a Sclerotinia forecasting model has identified key relationships between environmental conditions and Sclerotinia behaviour for UK isolates. This information has great potential to enable: i) prediction of when apothecia will appear and hence when ascospores are present and ii) prediction of lettuce infection. However, a preliminary forecasting model requires incorporation into the HDC approved decision support software MORPH4 (Methods of Research Practice in Horticulture), and testing in a commercial field situation to assess its potential for timing fungicide sprays for effective Sclerotinia disease control. Currently, a Defra project, 'Improving the uptake of simulative models in commercial horticulture (MORPH) (HH3814 SX, 4/2004 to 3/2009, Warwick-HRI) is also redeveloping MORPH and the new version, MORPH5 (Collier, 2007). It will include the results of further research into industry requirements, such as, ease of data input and model output integration with current management systems and key questions relating to different commodities.

The Defra project HH3215TFV (Clarkson *et al.*, 2005) aimed to produce a preliminary forecasting model based on the results of specific controlled environment (CE) experiments and field studies. The sclerotial germination and the ascospore infection phase of the life cycle of Sclerotinia were modelled separately, by Warwick HRI and ADAS, respectively. CE experiments at Warwick HRI showed that sclerotia germinated between 10 and 20°C, with intermediate rates of germination at temperatures in between, and little germination at 25°C. However, the duration of a cold 'conditioning phase' prior to sclerotia being placed in the appropriate conditions for germination had a major effect on germination times of sclerotia.

For example, with no cold conditioning, sclerotia took over 80 days to germinate at 18°C, but after 15 days conditioning at 4°C, sclerotia germinated in 40 days. In field experiments done by ADAS, monitoring germination of sclerotia buried at two week intervals during the growing season, it was shown that the rate of germination and the % of sclerotia which germinated was high for early burials, but much lower for burials from June onwards. The CE data was used to develop a model that successfully simulated this observed pattern of germination in the field. Significantly, the germination model included the process of conditioning identifying the need for low temperatures before rapid germination of sclerotia could subsequently occur. There was however some variability in the accuracy of the model in predicting exactly when apothecia were produced by the buried sclerotia in the field across years and field sites. For a very few burials this was as much as three weeks and this variability will be a concern as the model is developed in this project.

Modelling of Sclerotinia infection and disease, by ADAS was based on further CE data generated at Warwick HRI. Although spore germination was found to require \geq 97% RH *in vitro*, disease occurred in lettuce plants maintained at 50% RH with increasing rates of infection up to 100% RH. Temperature also affected the rate of disease increase, with slow development at 7°C and the quickest rate at 25°C. No disease occurred at 30°C. The infection model accounts for the interaction between temperature and RH effects, and was validated using data from ADAS field experiments, using results of monitoring natural infection and inoculating lettuce with ascospore suspensions. The model predicts the onset of disease and also the final % of lettuce infected if disease levels are relatively high, but there is more uncertainty in predicting low levels of disease.

The potential to forecast Sclerotinia disease is also being investigated in carrots in the current HDC project, 'FV 260 - Carrots: forecasting and integrated control of Sclerotinia disease'. A simple forecasting system is being developed, based on crop growth stage and environmental factors. From the first two years report, 1 Mar 2004 – 1 Mar 2006, canopy closure, the presence of fruiting bodies (apothecia) and senescing leaves on the ground were found to be the important factors affecting initiation and development of Sclerotinia disease in carrot crops. The most effective foliar fungicides were Signum and Shirlan (fluazinam), and it was important to apply these early in crop growth, before canopy closure. Contans did not give control of foliar Sclerotinia disease, but further experiments are needed to investigate whether it had an effect on sclerotial survival in the following winter.

The results of multiple burials of sclerotia carried out under the Defra project HH3215TFV (Clarkson *et al.,* 2005) suggested that successive flushes of apothecia could occur during

the season. Whilst it was thought that cold conditioning could occur at any soil depth, it has been shown that the germination phase requires the sclerotia to be close to the soil surface in order to produce spore releasing apothecia. We have inferred from experiments in the previous project that the early flushes of apothecia are therefore likely to be associated with sclerotia left on the soil surface or brought to the surface by cultivation in the previous autumn. These sclerotia then condition over the winter and germinate when temperatures rise in early spring.

However secondary flushes may also be initiated when soil is cultivated prior to planting in the spring when conditioned sclerotia may be brought to the surface to begin the germination process. However, sclerotia formed from diseased crops early in the season are unlikely to produce apothecia in the same season because the low temperatures required for conditioning do not occur. Prediction of the first flushes of apothecia appearing in the spring is therefore important either to optimise the time to spray a lettuce crop or to support a decision not to spray and thus reduce costs.

A major outcome of this project will be the development of a Sclerotinia forecasting model that can be used by growers. It will be necessary to reflect some of the model uncertainty in the way the prediction of the first apothecial flushes is presented. It is also imperative that the data requirements are limited to those available to growers and do not require an excessive number of additional inputs which could lead to frustration and rejection of a system. The MORPH project team have conducted interviews and surveys of end user needs and this information will be used when developing the model presentation for the end user (Defra project HH3814 SX). The main benefit to the industry of having a forecasting system for Sclerotinia control in lettuce is the potential to reduce the number of fungicide treatments or to justify treatments when needed. In addition, a forecasting system could result in improved efficacy through better timing of fungicide applications. Improved control of Sclerotinia through a comparison of fungicide products, combined with development of a Sclerotinia forecasting model, would result in fewer losses for the industry. A 10% loss in the national lettuce crop area due to Sclerotinia equates to a loss in production worth £4.2 million (Defra Horticultural Statistics 2004). Furthermore, some results from this project may also be applicable to other horticultural and agricultural crops susceptible to Sclerotinia, such as carrots, celery, parsnips, brassicas and oilseed rape.

The number of fungicides approved for use on lettuce against Sclerotinia is limited, and therefore there is interest in current tests on efficacy of fungicides against Sclerotinia and other common diseases, including downy mildew and Botrytis. Rovral (iprodione) and Amistar (azoxystrobin) are currently approved for control of Sclerotinia on lettuce (SOLA nos. 2004/0513 and 2001/1465 respectively). Signum (boscalid + pyraclostrobin) has full label approval for Sclerotinia control on outdoor lettuce. These act by killing the ascospores released by apothecia. Fungicides of particular interest at present for inclusion in experiments to compare efficacy are Amistar, Signum, Switch, Rovral, Thianosan (thiram), Octave (prochloraz) Teldor (fenhexamid). In addition, growers are interested in the biological control product Contans. This is a formulation of the mycoparasite *Coniothyrium minitans*, which colonises sclerotia of *S. sclerotiorum* in soil, causing them to become non-viable, and has good potential to control Sclerotinia in field grown lettuce. However, to be effective, Contans needs to be applied to soils at least 3 months prior to planting. This is not possible for many lettuce growers who use rented land, which is not accessible until immediately before planting. It was not included in the current experiment because both sites used were on rented fields and there was no access to the main field area before planting.

The objectives of this project are as follows:

- 1. Develop a Sclerotinia disease forecasting model and incorporate it into the disease forecasting system MORPH (developed at Warwick-HRI).
- 2. Evaluate the efficacy of Sclerotinia disease control on lettuce using fungicide spray applications timed according to the forecasting model.
- 3. Compare the efficacy of different fungicides and a biological control agent for Sclerotinia control.

Materials and methods

Development of the Sclerotinia forecasting model

In order to adapt the existing infection and germination models from a scientific tool into a forecasting tool, there were two key issues to consider. Firstly the structure of the sclerotia germination model was not suitable to run in forecasting mode as it is currently formulated to run with hindsight of the weather. Secondly, a major unknown for the infection model was estimations of spore numbers, which are difficult and costly to assess in the field and impractical for a forecasting system to be used by growers. Furthermore, it is not sensible to run an infection model without a good prediction of spore numbers, because there are many opportunities for infection to occur during the season as determined from previous work (Young *et al.*, 2004). In this project it was therefore decided to concentrate on predicting the

germination of sclerotia, using the assumption that if apothecia are present, so are spores in large numbers and infection is then highly likely. Sclerotinia spore germination and infection can occur over a wide range of temperatures and humidities, i.e., under almost all field conditions encountered during lettuce growing. Rather than reworking the sclerotial germination model from first principles, the existing published model for carpogenic germination of *S. sclerotiorum* sclerotia (Clarkson *et. al.*, 2007) was modified to run in forecasting mode and tested with data from this 2007 lettuce crop season.

Sclerotia germination model – initial derivation

The simulation model for germinating sclerotia (Clarkson *et al.*, 2007) forms the basis of the forecasting model. The original model simulates two phases; the first conditioning phase (equation 1) must be completed before the germination phase (equation 2) can proceed. Both phases of the model are driven by soil temperature and a soil water potential threshold was also imposed in each phase.

The model runs on a half hour time step with time series of soil temperature at 5 cm and a measure of water potential (kPa) measured at the monitoring site. Initially, at each time step the progress of the conditioning phase (equation 1) is calculated according to temperature provided the soil water potential is above the threshold. Over a series of time steps when the cumulation of equation 1 reaches unity, the sclerotia are fully conditioned and progress towards germination can then begin. Again this is calculated at each time step provided the soil water potential is not below the threshold and the results accumulated until the value reaches unity. At this time point, it is predicted that sclerotia have reached 50% germination (T50). This predicted value of 50% germination is then passed in to equation 3 which describes a cumulative germination curve from which other important measures such as the T10 (time to 10% germination) can be determined.

$$r_c = a + be^{-kT} \tag{1}$$

Where r_c is rate of conditioning per day, a, b and k are constants and T is temperature °C. Conditioning is set to zero above 20 °C and to a maximum at 4 °C.

$$r_{\sigma} = \exp(d_{\rho} + d_{1}/(T + 273)) \tag{2}$$

Where r_g is rate of germination per day; d_0 and d_1 are constants and T is temperature (°C). Germination rate is set to 0 for temperatures > 25 °C.

$$M = \exp(m + 0.5s^2) \tag{3}$$

Where M is the mean time to germination and m and s = 0.1417 are the mean and standard deviation of the corresponding normal distribution. Further details of the model derivation can be found in the original paper (Clarkson et al, 2006)

The germination model was originally fitted to data from two Sclerotinia isolates; isolate 13 which was used in a series of burials at a Cheshire field site and isolate TM which was monitored at a Norfolk site.

The model parameters for each isolate are tabulated below, Table 3.

S. sclerotiorur	<i>n</i> isolate 13	S. sclerotiorur	S. sclerotiorum isolate TM		
Parameter	Parameter	Parameter	Standard		
	value	Error	value	Error	
а	0.03273	0.00395	0.01056	0.001	
b	1000	*	1.28	1.61	
k	1.498	0.398	0.435	0.118	
d ₀	31.12	4.36	24.8	3.38	
d ₁	-10138	1236	-8422	961	

Table 3. Parameter values for conditioning and germination models (Clarkson et al., 2007).

Sclerotia germination model – modifications for forecasting

In order to adapt the model to operate in forecasting mode in this project, the following points needed to be addressed:

- 1. The water potential threshold should to be replaced by a surrogate function that could be calculated at any given site with the available data e.g. rainfall
- The model should be able to estimate T10 without the need to estimate T50 first. A measure of T10 is needed to give the grower sufficient warning of an oncoming flush of apothecia.

- 3. A lower temperature threshold for germination should be considered (equation 2) as sclerotia left close to the soil surface in the autumn could potentially germinate slowly over winter.
- 4. For simplicity and applicability, the model needs to be run using parameters for a single representative isolate, although in reality there are numerous isolates all with potentially different conditioning / germination rates.
- 5. The model needs to take account of increases in soil temperature that occur when a crop is fleeced.

Replacing soil water potential threshold with a threshold triggered by a temperature-rainfall function

Soil water potential thresholds in the original germination model were set in the range -4 to - 12.25 kPa for the two isolates at two sites monitored by Clarkson *et al.*, (2007) and the optimum threshold varied between each year of experiments even at the same site. Growers do not routinely monitor soil water potential, yet some measure of soil moisture is important to indicate when suitable conditions for germination are present. In order to run the model at any site, it was necessary to replace the soil water threshold, which enables germination to accumulate towards its goal or not at each time step. Therefore a function of rainfall and temperature was developed to replace water potential within the model as these data should be widely available at or close to a grower's site. This function was optimised based on previous data for the two sites presented in Clarkson *et al.*, (2007).

The simple function that replaces the water potential threshold is applied for all sites and in all years and is expressed as follows. If the temperature is < 12° C then soil moisture is not assumed to be limiting and germination can progress. Hence, during winter and early spring when soil temperatures are low, the soil is considered moist enough for germination to proceed. This is not unreasonable as many soils are close to field capacity around this time. If the average temperature in the preceding 24 hours is > 20° C then soil moisture is limiting and progress towards germination cannot accumulate during that time, this basically mimics hot conditions usually associated with low water potential which impede germination. If the average temperature is > 12° C and < 20° C in the preceding 24 hour then germination only progresses if there has been greater than 4 mm total rainfall in the previous 4 days. The performance of this new threshold based on temperature and rainfall is reported in Appendix 2. It is important to note that in the original model the water potential function is generic and therefore applicable to any field site and requires no local calibration.

Estimating the mean time to reach 10% sclerotial germination

The statistical germination model estimates the time to T50 using half-hourly sampled weather data for the season. At each time step equation 2 is calculated and when the cumulative reaches unity, the mean time to germination (T50) is said to occur, after which the lognormal distribution (equation 3) is fitted to give the cumulative germination curve. The main restriction of this approach for forecasting is that it would require weather data for several months in advance to predict T50 before the germination curve could be fitted and a T10 estimated. Clearly this is not feasible. One solution would be to initiate the model with weather data from the preceding season or an average of several seasons (depending on the data available to the grower), then as new weather data is available week on week this replaces the 'typical season' weather and so the model is re-run and a new prediction made.

A much more desirable solution would be to predict T10 in a different way, so that less weather data is required in advance, and as such the new season weather data would be more informative in the model. This is possible by taking advantage of the original model formulation where the ratio of T50 to T10 for each of the burial dates at the two sites reported in Clarkson *et al.*, (2007) is a constant ratio of 0.833. This ratio was used to estimate T10 by multiplying the germination equation 2 at each time step. This enables the model to calculate T10 whilst using significantly less weather data in steps ahead before it can be updated with the current season's weather. A prediction of T10 is an indication of the onset of a flush of apothecia. How this date should best be interpreted to inform decision making with regards to fungicide sprays is considered in the discussion section.

Germination Temperature Threshold

Clarkson *et al.*, (2007) demonstrated that germination can still occur; albeit very slowly at 5 °C. Freezing temperatures tend to damage sclerotia and hence apothecia are not produced at all. Germination at temperatures below 5 °C was thought to be so slow that it was insignificant (pers. comm., J. Clarkson 2007). However, if viable sclerotia are buried in the early autumn the cumulative affect of many cold days may progress germination significantly through the winter and hence a lower temperature threshold should be imposed. A threshold of 4 °C below which progress towards germination could no longer occur was added to the model and tested against some of the published data. In general for later spring burials this additional threshold makes no difference to the modelled time to germinate. For the burial on 17 December 2003 in Norfolk (Clarkson *et al.*, 2007) the threshold had the effect of delaying the predicted T10 by 17 days compared to the original model which resulted in a better model fit.

Different isolates of S. sclerotiorum

Conditioning and subsequent germination rates at certain temperatures may vary between Sclerotinia isolates. Clarkson *et al.* (2007) fitted the original germination model with data for two Sclerotinia isolates (13 and TM) and found that isolate 13 was consistently faster both for conditioning and germination phases at a given temperature. Under field conditions, isolate 13 germinated significantly faster than TM (Figure 5). Provisional results from a current Defra project (HH3230SFV) have suggested that there are many Sclerotinia isolates present at a given field site with variable conditioning and germination rates, but that isolate 13 was consistently one of the fastest to germinate. Growers have no means of knowing the germination potential of local isolates so it was decided that the model would use parameters associated with isolate 13 in the model to predict germination of sclerotia as this would provide an indication of the earliest time an apothecial flush could occur and spores released.

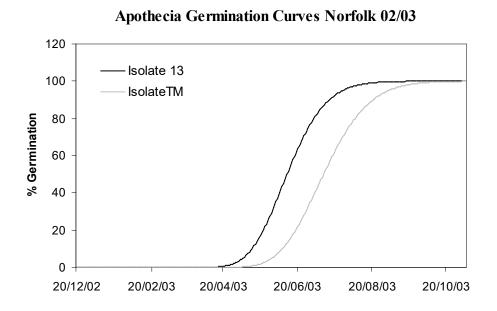


Figure 5. Modelled germination curves for two sclerotia isolates, 13 and TM, both buried on December 20th 2002.

Fleeced Crops

Due to the premiums associated with early lettuce crops, growers may fleece their crop which raises both the air and soil temperature beneath. The action of raising soil temperature would decrease the time to germination of sclerotia bodies within the soil. The model therefore has had a simple function added to represent fleecing which is assumed to

cover the soil from the planting date until April 1st as a default. Data on raised soil temperature beneath fleeces is limited but a mean gain of 1.4°C per day in April is reported by Rickard (1978) and used as a default in the model throughout the diurnal cycle.

Model validation

New model predictions of T10 with the new rainfall-temperature function and the modification to predict T10 directly have been compared with the results published in Clarkson *et al.*, (2007) for burial dates at each site where data was available. Further details are reported in Appendix 1.

Model Summary and Data Requirements

To summarise, the forecasting model for germination of sclerotia:

- is a model based on the carpogenic germination model of Clarkson et al,, (2007).
- is adapted to forecast time to 10% germination of sclerotia (T10) as an indicator of the onset of a flush of sclerotia.
- is adapted to operate with routinely collected field data on a half-hourly basis.
- has comparative predictive capability to the original simulation model.
- uses historical local weather data in the forecast which is updated as the current season's data becomes available.
- includes an option for pre-conditioned and unconditioned sclerotia.
- includes an option to simulate fleeced crops.
- is parameterised using a fast germinating isolate to indicate the earliest production of apothecia.

The model requires time series of weather data: to include temperature at 5cm soil depth (or air temperature if unavailable) and rainfall data at half hourly intervals. Data at hourly intervals can readily be used and daily data can provide an approximate hourly series if a local hourly recording logger is unavailable.

The user will also need to consider a start date for the germination model to start. This is straightforward if applied to sclerotia produced in the laboratory and buried in the field. However, for natural sclerotia in the field, the dates of tillage operations will be required as this is when sclerotia will be brought to the soil surface ready to germinate. In this scenario, many of the sclerotia may already be fully conditioned if they have been in the soil over winter and the model allows this phase to be by-passed if required. The importance of these tillage dates and an option of using a default date during the autumn are considered in the discussion section of this document.

Field site details

There were two field sites, located in growers crops in Cambridgeshire, negotiated with the assistance of consultant David Norman. As is normal practice, the fields selected were on rented land, and therefore there was no access for any field treatments prior to lettuce planting.

Site 1: Merrymac Salads, Mudds Drove, Three Holes, Wisbech, Cambs PE14 9JU, Buckinghams field (working area 12.17 ha), OS GR TL 672 897, nr Feltwell.

The previous crop was winter wheat, with no cultivations after harvest until ploughing immediately prior to lettuce planting in March 2007. Lettuce cv. Edition were planted as blocks on 15 March (4-5 true leaves) and harvested 16 May. The soil type was peat/organic, in excess of 40 % organic matter. The crop was under non-woven fleece (Gromax Industries 17g/m2 (spunbonded filaments) 100% polypropoline) for the whole duration.

Site 2: G's; J B Shropshire & Sons, Hainey Farm, Barway, Ely, Cambs CB7 STZ, field CW63 (working area 15.46 ha), OS GR TL 630 792, Lark Bank, nr Prickwillow.

The previous crop was winter wheat, with no cultivations after harvest until ploughing immediately prior to lettuce planting in March 2007. Lettuce cv. Saladin (4-5 true leaves, Scotts compost Lev B2 Bulk, 4.0 cm³ blocks) were planted on 1 March 2007 and harvested 10 May. The crop was under non-woven fleece (17g, Lows of Dundee, UK) for the whole duration and was irrigated by overhead boom once on 10 March 2007, with 15mm water.

Sclerotinia sclerotiorum isolates and production of sclerotia

The isolates used in this project were all derived from Sclerotinia infected lettuce plants and are coded as follows:

Isolate 13: 'standard' Warwick HRI isolate, Cheshire, UK, 1996 Isolate TM: 'standard' Warwick HRI isolate, Norfolk, UK, 1996 Isolate HDC 1-1: from G's (fields between Southery and Methwold), 2006 Isolate HDC 2-6: from Merrymac Salads, Feltwell, 2006 Isolates 13 and TM were used to derive the original carpogenic germination simulation model for *S. sclerotiorum* sclerotia (Clarkson *et al.*, 2004). Isolates HDC 1-1 and 2-6 were 'local' isolates to the field experiment sites in this project. All isolates were different from each other as indicated by incompatibility in mycelial compatibility tests carried out according to Schafer & Kohn (2006).

Original isolations were made by surface sterilising the sclerotia from infected lettuce in 50% v/v sodium hypochlorite and 70% ethanol for 4 min with agitation, followed by two washes in sterile distilled water (SDW) for 1 min. Sclerotia were then bisected, placed on potato dextrose agar (PDA; Oxoid) and incubated for 4 weeks at 20°C. Sclerotia formed in culture were then removed, stored at 5°C and used as a stock supply for all further cultures.

To produce large numbers of sclerotia for the field experiments, two agar plugs (approx. 2 mm²) from the edge of four-day-old *S. sclerotiorum* colonies derived from stock sclerotia were used to inoculate sterile wheat grain (25 g, wheat grain, 80 g water autoclaved at 121 °C for 15 min) in 500 ml conical flasks. Flasks were incubated at 20°C and shaken gently by hand twice a week to encourage formation of uniform sclerotia and prevent clumping of wheat grain and mycelium. Mature sclerotia were formed after approx. four weeks and harvested by wet sieving to recover those between 2 and 5 mm while the wheat grain was floated off. Finally the sclerotia were dried in an air-flow cabinet overnight after which they were ready for use.

Weather data loggers for field experiments

A Delta-T weather logger was located as close as possible to the proposed field sites on 21 December 2006. For G's, the logger was sited in the field used for the experiment the following spring. At Merrymac Salads, the logger was initially located at Manor Fen Farm near Feltwell, and moved to the lettuce field on the day the plots were set up (16 March 2007). The weather loggers were set up at the edge of each experiment area, with wires to probes such that probes were located within or between plots as appropriate. All the probes except the rain gauges were positioned under the fleeced crop; therefore no adjustment was necessary in the model for raised temperatures under fleece. Each logger recorded the following variables at hourly intervals: mm rain, soil temperature °C at 2 cm depth (5 probes),

air temperature °C, % relative humidity, leaf wetness as % of time wet (two sensors, one horizontal and one angled). Data was downloaded and checked weekly. Probes and logger batteries were checked and replaced if necessary. Weather data from the growers loggers at both G's and Merrymac was also supplied to ADAS for September - December 2006, for use in developing the forecasting model.

Sclerotinia inoculum for field experiments

Before sclerotia were used in the field experiments they were buried at a depth of 30 cm on 21 December 2006. This winter burial was to allow the sclerotia time to condition, a process which has been shown to occur at low temperature and which allows subsequent rapid germination when the soil temperature increases in spring (Clarkson et al., 2007). Sclerotia from all isolates were subsequently retrieved and re-buried in grids between the modelling and fungicide experiments at each of the two field sites, the day after lettuce were planted, at a depth of 1 cm (50 sclerotia x 3 reps per isolate). Grids were monitored weekly for appearance of apothecia so that germination times could be compared between isolates and also with the time predicted by the germination model.

A mixture of sclerotia from isolates HDC1-1 and HDC 2-6 were also used to inoculate all plots at each site, for each experiment, at the time of setting up the experiment plots (the day after lettuce planting), at a rate of approximately 100 sclerotia per plot. Sclerotia were scattered over plots and raked in to a depth of about 1cm around the lettuce plants.

Field experiments to evaluate the Sclerotinia forecasting model

Plots were marked out in the growers crops at G's and Merrymac sites (site descriptions in above section 'field sites') on the day following planting at each site. The crop dates were: G's: cv Saladin, planted 1 March, harvested 10 May. Merrymac: cv Edition, planted 15 March, harvested 16 May.

There were 5 treatments to the Sclerotinia forecasting model experiment ('modelling experiment'):

1. Untreated

- 2. Fungicide sprays applied as timed by the Sclerotinia model
- 3. Fungicide applied at spray time 1 (early in crop development)
- 4. Fungicide applied at spray time 1 & 2 (early and mid crop)
- 5. Fungicide applied at spray time 1, 2 & 3 (early, mid and late crop)

Signum (boscalid + pyraclostrobin) was the selected fungicide used for treatments 2, 3, 4 and 5, applied according to EPPO guidelines. The experiment design was a randomised complete block, with 5 treatments and 6 replicate plots per treatment (= 30 plots). Plot size was selected to ensure that there were at least 100 assessed lettuce each, with an unassessed buffer area allowed around each plot.

The forecasting model was run prior to planting based on weather data recorded at the site from 21 December 2006 and the model could predict for the weeks ahead using data for the site from the previous season. The model was run to predict both conditioning and germination using the burial date of 21 December 2006 as a start time to give the first estimate of T10 for the 'fast' sclerotia isolate 13. Each week post planting the weather data logger was downloaded and checked on the same day if possible, and the model re-run to predict the time to 10% sclerotial germination. A prediction of 10% germination of sclerotia one to two weeks ahead of the time of running the model would initiate an alert the ADAS site manager by email or telephone so that preparation for treatment sprays could be made. An alert for germination in the 2 weeks or so prior to the expected date of harvest would not result in a spray treatment because of minimum harvest interval requirements. For treatments 3, 4 and 5, the appropriate plots were sprayed with Signum, at weeks 2, 4 and 6.

Lettuce total diameter and heart diameter were measured on a subset of 5 randomly selected lettuce in one untreated plot each week (same plot each week), to monitor lettuce growth progress. Disease was assessed and recorded after 2 weeks, 4 weeks and then weekly, on all assessed plants per plot. Sclerotinia = 1 (diseased) or 0 (no disease), similarly for Botrytis. Downy mildew was recorded both as incidence and as % area affected. For spraying and assessing, the crop fleece was removed beforehand, by G's staff with prior arrangement, and by ADAS staff at the Merrymac site. Sclerotial germination was assessed weekly in all grids, recorded as the presence or absence of apothecia.

Field experiments to compare Sclerotinia control using different fungicides

Plots were marked out in the growers crops at G's and Merrymac sites (site descriptions in above section 'field sites') on the day following planting at each site, adjacent to the modelling experiment. The crop dates were:

G's: cv Saladin, planted 1 March, harvested 10 May.

Merrymac: cv Edition, planted 15 March, harvested 16 May.

The following fungicides were selected for inclusion in the fungicide experiment after consultation, in particular with consultant David Norman and Vivian Powell (HDC). There were 8 treatments in total, including an untreated control treatment (Tables 4 and 5).

Treatment number	Trade name	Active ingredient	Approval status for outdoor lettuce		
1	Untreated				
2	Amistar	(azoxystrobin)	SOLA 1465/01		
3	Signum	(boscalid + pyraclostrobin)	Approved, for sprays 1 April to 31 Oct		
4	Switch	(cyprodinil + fludioxonil)	SOLA 2079/07, max. 2 treatments per crop		
5	Rovral Flo	(iprodione)	SOLA 0513/04		
6	Bayer (new) UKA383	N/a	Not approved		
7	Octave	(prochloraz)	SOLA 0650/01		
8	Teldor	(fenhexamid)	SOLA 0026/05		

Table 4. Fungicides, active ingredients and approval status

A three-spray programme (Table 5) was selected for all fungicides, regardless of label restrictions, to ensure comparability between products, timed to ensure a sufficient minimum harvest interval for an early crop where the duration of the crop could only be estimated.

Trt		We	Weeks post planting*								
		1	2	3	4	5	6	7	8	Rate	Water vol/ha
1	Untreated										
2	Amistar		х		х		х			1 L/ha	400
3	Signum		х		х		х			1.5 Kg/ha	400
4	Switch		х		х		х			0.8 Kg/ha	400

5	Rovral	х	х	x	2.3L/ha	400
6	Bayer (new) UKA383	x	x	x	0.5L/ha	400
7	Octave	х	х	x	200g/ha	400
8	Teldor	х	х	x	1.5 Kg/ha	600

* The spray dates for both sites were 27 March, 12 April and 25 April.

Spraying at G's was originally planned to begin two weeks earlier but was delayed due to bad weather.

The experiment design was a randomised complete block, with 8 treatments and 5 replicate plots per treatment (= 40 plots). Plot size was selected to ensure that there were at least 100 assessed lettuce each, with an un-assessed buffer area allowed around each plot. Sclerotia were spread evenly over each plot after planting and raked around lettuce.

Disease was assessed as for the modelling experiment, two weeks after each spray, and once more immediately prior to harvesting. Phytotoxicity assessments on each plot were made two weeks after each spray, following EPPO guidelines (PP1/135(2) Phytotoxicity assessments). Prior to field harvest, a subset of 25 untrimmed lettuces per plot was weighed, then trimmed and weighed again. The 25 lettuce were selected to be in adjacent rows in the middle of each plot, and if a lettuce was missing due to previous Sclerotinia infection, another lettuce was chosen such that 25 lettuce were weighed (the numbers of missing lettuce were recorded).

Results

All treatments and assessments were made as planned at both sites. The sites differed in the diseases observed, eg., no downy mildew was recorded at Merrymac, and no Sclerotinia disease at G's (Table 6).

Table 6. Summary of data collected from field work: modelling experiment and fungicide experiment

	Modellin	ig experiment	Fungicide experiment			
	Merrymac	G's	Merrymac	G's		
Germination of sclerotia in grids	*Yes	*Yes				
Lettuce growth stage	Yes	Yes	Not done	Not done		
Sclerotinia disease	Yes	No Sclerotinia	Yes	No Sclerotinia		
Downy mildew (DM) severity	No DM	Yes	No DM	Yes		
Botrytis	Not done	Not done	Yes	Yes		
Lettuce weight	Not done	Not done	Yes	Yes		

* sclerotia in grids were located between the modelling and the fungicide experiment.

Field experiments to evaluate the Sclerotinia forecasting model

The dates of planting and last assessment for the modelling experiment at the two sites are as follows:

G's: planted 1 March; first germination (grids) 7 May, harvest 10 May, cv Saladin.

Merrymac: planted 15 March; first germination (grids) 1 June, 'harvest' 1 June, cv. Edition (note: plants were allowed to go beyond the harvest date for yield (16 May for fungicide experiment), to enable monitoring of sclerotial germination for as long as the field was available to the experiment.

Predictions of time to reach 10% sclerotial germination (T10)

The model was validated against data collected at the Merrymacs site during the spring of 2007. Where lettuce were irrigated aerially the monitored rainfall included capture of any irrigated water. The model was run for a half-hour sampling interval with start date of 21st December. The first model predictions were initiated with 2006 weather data from ADAS

Terrington site (30km from the Merrymac site and 49km from the G's site). As the weeks passed the data recorded from the Merrymacs site replaced old data, and new predictions of T10 were continually updated. The temperature series at Merrymacs (Figure 6) included the 30cm depth measure followed by the 5cm depth measure to replicate the conditions that the sclerotia had encountered.

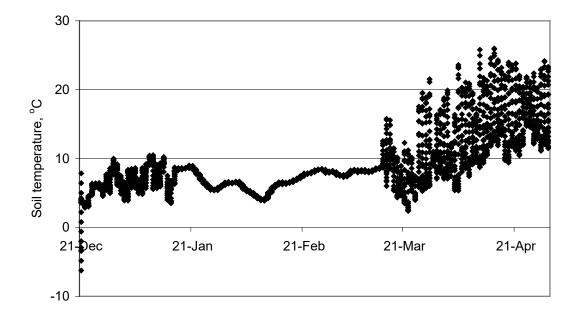


Figure 6. 2007, Merrymacs: soil temperature data used in the Sclerotinia forecasting model. From 21st December to15th March the sclerotia were buried at 30cm, hence the damped temperatures observed. After 15th March temperatures were recorded at 5cm depth (minimum depth possible for probes) when the sclerotia were reburied at 1cm depth.

Model runs made during mid-April indicated that the T10 of the apothecia would occur at the end of April at G's and early May for Merrymacs. This information was used to decide when and *if* to apply fungicide treatments. During April, with the occurrence of very dry weather at Merrymacs (only 0.9 mm rainfall in April) (Figure 6) this date was pushed back further thus making a decision not to apply any sprays as part of treatment 2 at Merrymacs. By mid May the lettuce crop was ready for harvest. Given weather data up until the end of May 2007, the model estimated that the T10 would be on May 18th.

At the Gs site, the model was initiated with weather data for Terrington 2006 and updated with site data as it became available, though temperatures at both sites are similar as would be expected due to their close proximity. Initial predictions of T10's were similar to

Merrymacs as would be expected but model runs made during early April predicted a T10 several weeks in advance of Merrymacs i.e., during mid-April. As a result the first application for Treatment 2 (sprays applied according to model predictions) was put on the G's modeling experiment on 25 April. During the early part of the dry April, G's site had more rainfall events and had also been irrigated on 15 March 2007 with 15 mm (no irrigation was done at Merrymacs). In particular there was a significant rainfall event of 16mm at G's on April 11th which Merrymacs did not receive. However, in late April, rainfall patterns were more similar at both sites (Figure 7). Each week during April when the model was run, the overall dry spell served to delay the predicted T10 further. However, the localised rainfall recorded at G's is a major factor in the final prediction of T10 (made with weather data up to the point of harvest) of May 1st, which is 17 days in advance of predictions of 18 May for apothecia at Merrymacs. Though, interestingly, total rainfall (21 December 2006 to 21 May 2007) was very similar for both sites, at 165 mm and 162 mm for Merrymac and G's, Clearly, localised rainfall events are able to affect the germination time respectively. significantly for sites with very similar growing season temperatures and the model has successfully simulated this behaviour.

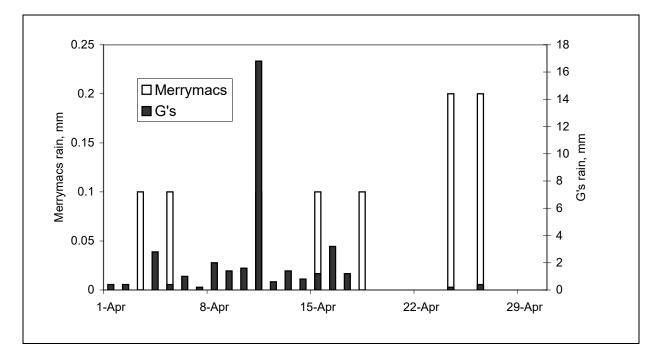


Figure 7. April 2007, rainfall at Merrymacs and G's farm sites. Note: Merrymacs had only 6 days of rainfall, each with 0.2mm or less recorded.

Sclerotial germination in grids

For sclerotia buried in grids on site in December 2006, germination started to occur towards harvest time, at both sites. Germination was first observed at G's on 7 May 2007 (isolates 1-1, 2-6, TM and 13 had 2, 0, 2.7 and 4% germination, respectively). Germination was first

observed at Merrymac on 1 June 2007 (isolates 1-1, 2-6, TM and 13 d 6.7, 7.3, 16.0 and 32% germination, respectively). This compares well with the model results which predicted that the T10's of the sclerotia for isolate 13 would occur on 1 May 2007 at G's and 18 May 2007 at Merrymac.

Diseases in the modelling experiments

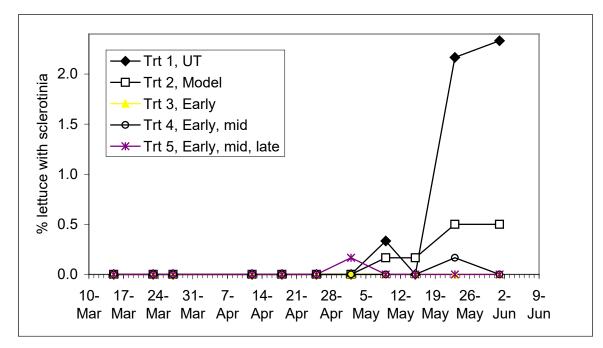


Figure 8. Merrymac 2007, modelling experiment, % lettuce with Sclerotinia disease, cv. Edition (F = 0.08). Trt 2, Model, was untreated (i.e., no spray alerts according to forecasting model); commercial harvest date was 16 May; apothecia were first observed 1 June.

Sclerotinia disease was observed at low incidence at Merrymac, late in the experiment (Figure 8), with most disease seen in untreated plots, and lowest disease in the plots sprayed three times (Trt 5), but overall the difference between treatments for Sclerotinia incidence was not significant. The model did not give an alert for the need to spray fungicides (Table 6), and therefore Trt 2 ('Model') was equivalent to Trt 1 (untreated).

No Sclerotinia disease was observed at G's in the untreated plots (Table 6) or any other plots of the modelling experiment, despite there being some apothecia germination, first seen 7 May.

Table 6. 2007, site summary for Sclerotinia modelling experiment.

	G's site	Merrymac site
Planting date	1 March	15 March
Fleece on crop (whole duration)	Yes	Yes
Harvest date (for yield)	10 May	16 May
Rain, 21 Dec '06 to 21 May '07	162	165
Irrigation	Yes	No
Date first apothecia seen in grids	7 May (max. 4%)*	1 June, (max. 32%)*
Final prediction date for 10% sclerotial germination (T10)	1 May	18 May
Spray alert	Yes (sprayed 25 April)	No
Sclerotinia disease at harvest in untreated plots	0	<0.5%**

* % out of 150 sclerotia buried in grids

** Sclerotinia disease was assessed at Merrymac up to 1 June, i.e., beyond the commercial harvest date. On 1 June, untreated plots had an average of 2.3% Sclerotinia incidence.

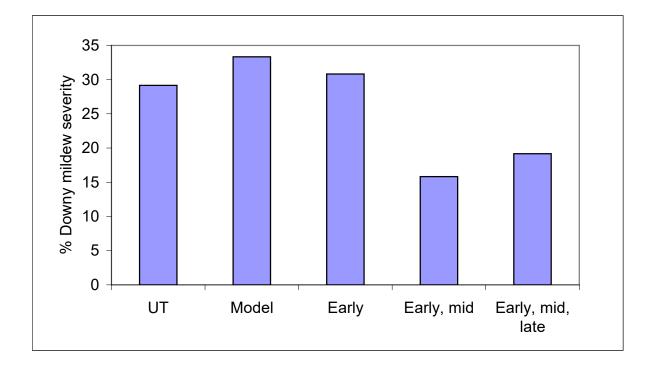


Figure 9. G's 2007, modelling experiment, downy mildew severity, cv. Saladin, 25 April (F = 0.26).

Downy mildew was severe at the G's site, with 100% incidence at all assessment times. Severity by 25 April ranged from 15-33% (Figure 9). Overall, there was no significant difference in downy mildew severity between treatments, but the two and three spray programmes had the lowest disease.

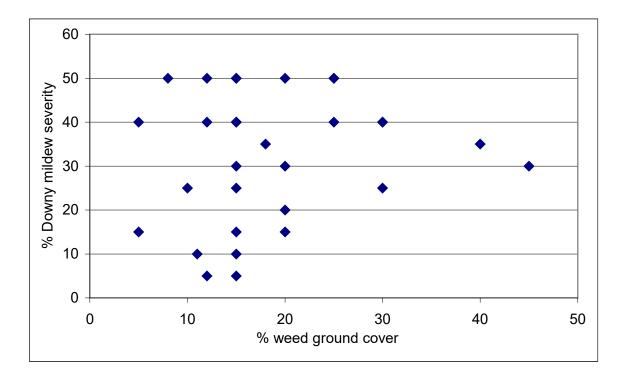


Figure 10. G's 2007, modelling experiment, downy mildew severity and % weed cover, cv. Saladin, 2 May (weeds: F = 0.93, downy mildew: F = 0.20).

The weed cover, almost all of which was Fat Hen, was high at G's, with more than 40% ground cover in some plots (Figure 10). There was no clear relationship between downy mildew severity and % ground cover with weeds.

Field experiments to compare Sclerotinia control using different fungicides

The dates of planting and harvest at the two sites are as follows: **G's**: planted 1 March; first germination (grids) 7 May, harvest 10 May, cv Saladin. **Merrymac**: planted 15 March; first germination (grids) 1 June, harvest 16 May, cv. Edition.

Progress of lettuce growth

The rate of increase in lettuce plant diameter was similar at G's (cv. Saladin) and Merrymac (cv Edition) (Figure. 11), but total diameter by harvest was larger at Merrymac (Figure 12).

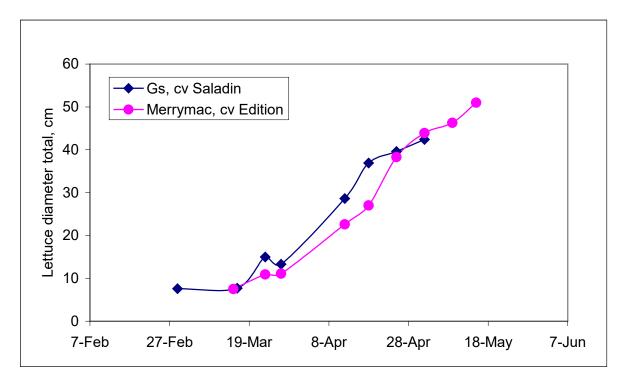


Figure 11 . Merrymac and G's, 2007, total lettuce diameter.

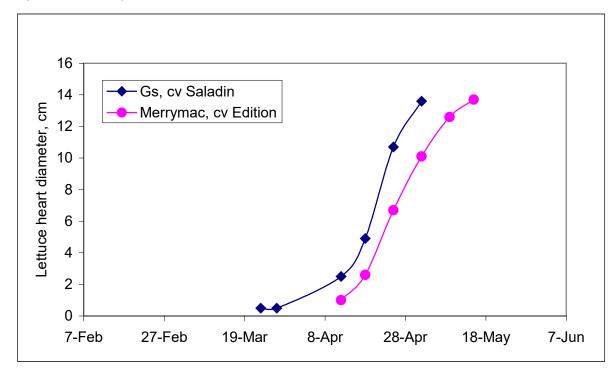


Figure 12. Merrymac and G's, 2007, lettuce heart diameter.

The rate of increase in heart diameter was similar at both sites (Figure 8), with the time difference reflecting the earlier planting date (G's planted 1 March, Merrymac planted 15 March).

Sclerotinia disease

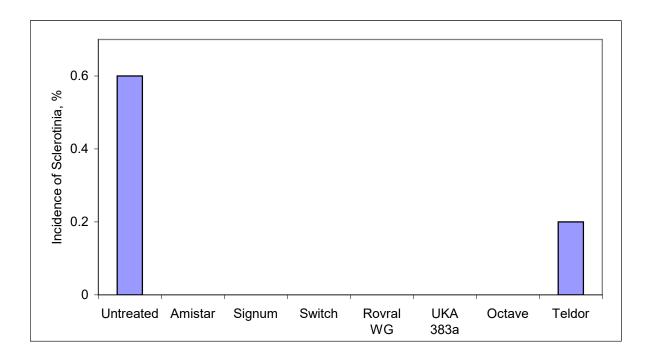


Figure 13. Merrymac 2007 fungicide experiment, % Sclerotinia disease, cv Edition, 16 May (F = 0.45).

Sclerotinia disease was only observed at Merrymac on cv Edition, late in the crop on 16 May (Figure 13). Only the Teldor treated plots and untreated plots had Sclerotinia disease, at low incidence (average of 0.6% or less). There was no Sclerotinia disease seen on cv Edition at the G's fungicide experiment site in 2007.

Downy mildew

Downy mildew was severe at the G's site, with 100% incidence at all assessments. Severity was 18% in untreated plots and UKA 383a treated plots on 25 April (Figure 14), and 10% or less for the other treatments.

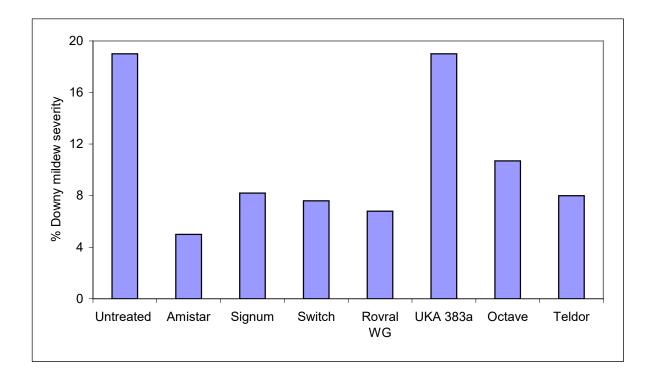


Figure 14. G's 2007 fungicide experiment, % downy mildew severity, 25 April, cv Saladin (F = 0.15).

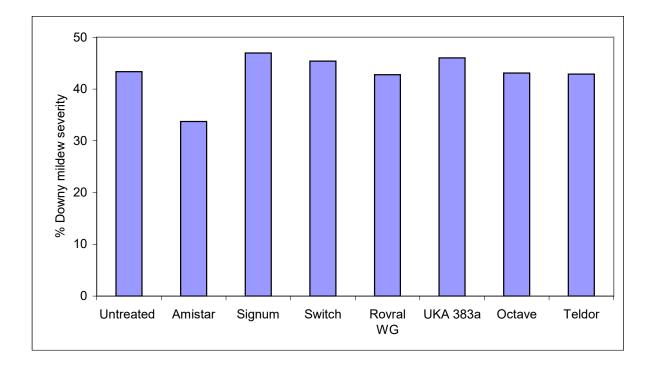


Figure 15. G's 2007 fungicide experiment, % downy mildew severity, 10 May, cv Saladin (F = 0.047).

By 10 May, all plants had 35% or more leaf area affected by downy mildew, but with significant differences between treatments (Figure 15).No downy mildew infection was observed at Merrymac (cv Edition).

Botrytis

By harvest at Merrymac, there were significant differences between fungicide treatments for Botrytis incidence, with Switch treated plants having no Botrytis, compared to 4% in untreated plots (Figure 16). No Botrytis was observed at G's in 2007.

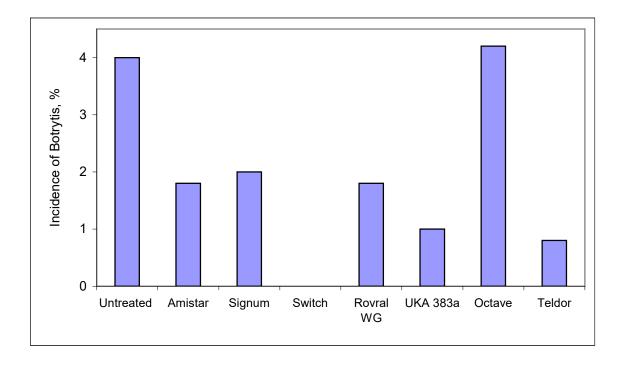


Figure 16. Merrymac 2007 fungicide experiment, % Botrytis incidence, cv Edition, 16 May (F = 0.002, SED trt = 0.98, LSD trt = 2.01).

Lettuce yields

There were significant differences in untrimmed weights (Figure 17) and trimmed weights (Figure 18) for lettuce at the Merrymac site. Switch treated plots had the lowest weight. At Merrymac the trimmed lettuce weights ranged from 0.55 to 0.72 Kg, but with no significant differences between treatments (Figure 15).

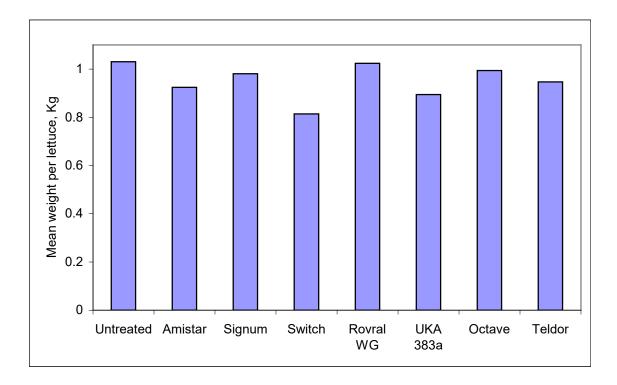


Figure 17. Merrymac 2007 fungicide experiment, untrimmed weight per lettuce, cv Edition, 16 May (F = 0.005, SED trt = 0.053, SED trt = 0.108).

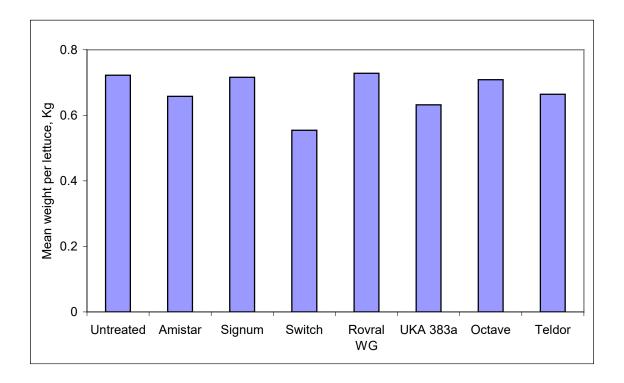


Figure 18. Merrymac 2007 fungicide experiment, trimmed weight per lettuce, cv Edition, 16 May (F = 0.002, SED trt = 0.04, LSD trt = 0.081).

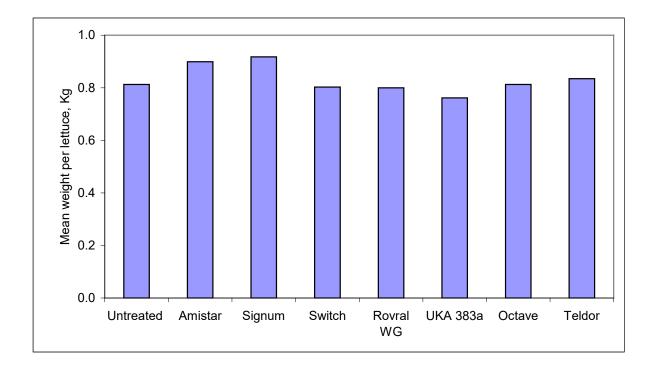


Figure 19. G's 2007 fungicide experiment, untrimmed weight per lettuce, cv Saladin, 10 May (F = 0.18).

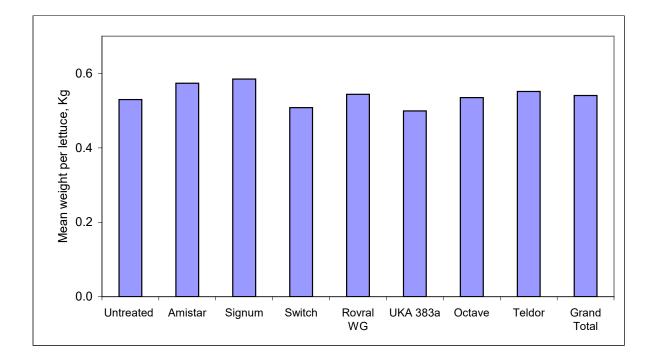


Figure 20. G's 2007 fungicide experiment, trimmed weight per lettuce, cv Saladin, 10 May (F = 0.46).

There were no obvious phytoxicity symptoms (e.g., leaf scorch or browning) in any of the fungicide treated plots at Merrymac in 2007, but it was noticeable that all plots treated with Switch had plant stunting, with plants not meeting between rows as in all the other treatments. This is reflected in the lower weights recorded for lettuce treated with Switch.

Unlike Merrymac, at G's there were no significant differences overall in untrimmed weight (Figure 19) or trimmed weight (Figure 20), (trimmed weight range was 0.50 to 0.58 Kg), but the lowest weights were from Switch and UKA 383a treated plots.

As at Merrymac there were no phytotoxicity symptoms such as leaf scorch, but Switchtreated plants appeared smaller. This was confirmed by measurements on representative plants on 25 April 2007, where the mean total plant diameter on untreated and Switch treated plants was 38.3 cm and 31.7 cm, respectively. The mean plant heart diameter on untreated and Switch-treated plants was 6.7 cm and 5.8 cm, respectively. There were no phytotoxicity symptoms observed on any of the treatments at G's.

Discussion

Sclerotinia forecasting model

This is the first attempt to develop and field-test a practical predictive model for germination of sclerotia from *S. sclerotiorum*, based on previous data from field experiments and controlled environment experiments (Clarkson *et al.*, 2004 & 2007). Predictions of the germination times of the buried sclerotia by the forecasting model were within the range of expected variability when considering the data on which the model was based (- 6 days for G's and -13 days for Merrymac). Spray decisions will need to take account of the fact that apothecia could develop several days before or after the prediction date given by the model. Results from the second year of this project will be needed to decide if the model will work for forecasting Sclerotinia disease.

The forecasting model for Sclerotinia in lettuce runs on fairly simple weather data, recorded at half hour intervals, but could be adapted to run on hourly intervals or even daily data, but with some increase in variability of predictions with daily data. The data can be obtained from regional weather loggers, but rainfall in particular can be localised, and an on-site rain gauge would be desirable. In-field temperature probes are desirable, because not only would they be more accurate for the field in question, but they can be placed to record temperatures under fleece, if fleece is used. If regional or local air temperature data is used, the forecasting model does have an option to adjust for the increased temperatures that occur under fleece. Given the variability associated with prediction times for sclerotial germination, the chances of success with the forecasting model would be increased by using on-site weather loggers. The exact requirements for MORPH will be confirmed in the second year of the project, to ensure that the model outputs are appropriate for growers and for MORPH. The forecasting model is planned to be programmed into MORPH by Warwick-HRI staff.

At the Merrymac site, the first apothecia were seen on 1 June, and the final predictions for the first apothecia were for 18 May for the fastest isolate of *S. sclerotiorum* (isolate 13). However, the commercial harvest date at Merrymacs was 16 May, too early for lettuce to have become diseased from the sclerotia buried in the grids and plots. At Merrymacs, Sclerotinia inoculum was present, but disease pressure was low overall and most likely the ascospore inoculum was produced too late to cause serious losses by harvest. The forecasting model did not give an alert to apply any fungicides for Sclerotinia. The three-spray Signum programme gave the best control, with no Sclerotinia disease, but disease pressure was low which probably explains why the early spray alone, and the early + mid-crop spray treatment also gave good control (<0.2% Sclerotinia incidence, compared to 2.3% in untreated plots).

The source of spore inoculum that caused lettuce infection in the experiment at Merrymacs is not known for certain, but was probably not from the any of the sclerotia used in the grids or scattered in the plots as inoculum, because apothecia in the grids were not observed until after disease was observed on lettuce plants. It is more likely that disease was due to natural sclerotia already in the field. To test this, sclerotia were retrieved from four infected plants and surface sterilised as before to produce cultures on PDA. The new isolates were then tested for mycelial compatibility with isolates HDC 1-1 and 2-6 which had been used to inoculate the field plots. All four isolates were incompatible with 2-6 and three were incompatible with 1-1 confirming that at least three of the isolates from the infected plants were different from those used to inoculate the plots. Although one of the new isolates was compatible with HDC 1-1, this may not necessarily be identical as several isolates can belong to the same mycelial compatibility group.

At the G's site the first apothecia were seen 7 May, which ties in well with the prediction of T10 on 1 May. The forecasting model triggered a Signum spray for 25 April. But by harvest on 10 May, no disease was seen in the crop in the untreated plots, despite 32% germination observed for isolate 13 on 7 May. Most likely, the germination of sclerotia occurred too late

to initiate visible Sclerotinia disease before harvest. The irrigation on 10 March with 15mm water was a very high proportion of the total water or rain in April at G's, and caused the prediction date for first apothecia to be revised from mid May to May 1st. In addition to the irrigation, there was significantly more rainfall during April at G's than at Merrymacs, a probable cause of higher % germination noted in the grids at each site. Low Sclerotinia disease pressure at the G's site has made it difficult to make good comparisons of the treatment results.

In this project, where the start date was taken to be the date of burial of sclerotia in December 2006, the forecasting model was run weekly, using new weather data for the most recent week, and all previous weather data from 21 December 2006 onwards. In practice, the forecasting model will most likely need to be run using at least two start dates, which are [1] the date of the last cultivation the previous year, e.g., harvest of a previous crop such as winter wheat, and [2] the ploughing immediately before lettuce planting the following spring. The first apothecia are most likely to be produced from sclerotia which have been brought to the optimum soil depth in the previous year. However, there may be later 'flushes' of apothecia produced from sclerotia which are brought to the optimum soil depth during the cultivations at the time of lettuce planting. Hence the forecasting model will need to be run with start dates of each cultivation time, to predict any further development of apothecia.

While it is possible to predict germination times of sclerotial germination, it is very difficult to make predictions of the concentrations of airborne Sclerotinia ascospores that will infect the lettuce and cause disease. Originally, this project aimed to combine the sclerotial germination and ascospore infection models as produced in the Defra project HH3215TFV (Clarkson *et al.*, 2005). However, the Sclerotinia infection model relies on estimating the concentration of ascospores in the air, and this is only possible if the number of viable sclerotia in the soil can be estimated. We can predict time to 10% germination of sclerotia, but without some method to estimate the number of sclerotia in lettuce fields and surrounding fields, we cannot know if 10% germination is from tens, thousands or more viable sclerotia at the right depth per unit area of soil. It may be possible in the future to measure spores in the air, if simple and inexpensive spore traps are developed for Sclerotinia.

Sclerotinia, downy mildew and Botrytis control with different fungicides

Comparison of Sclerotinia control with different fungicides was difficult because of the low levels of Sclerotinia disease. There was no Sclerotinia observed at G's in the experiment area, but scattered Sclerotinia-affected lettuce were noted in a low lying area of the field by G's staff.

At Merrymacs, Sclerotinia disease was only observed in plots treated with Teldor and in untreated plots, but with high variability between plots for numbers of plants affected by Sclerotinia, the treatment differences were not significant.

Downy mildew did not occur at Merrymacs, probably due to a combination of cultivar (Edition) and low inoculum. However, at G's, cv Saladin, downy mildew was severe, with 100% incidence at all assessment times, and high severity. On the 25 April, severity was 18% on the untreated plants, By 25 April, all the fungicides except UKA 383a (which had similar severity to the untreated, 18%) gave approximately 50% control or better. However, by harvest, all treatments had 30% or more severity, with relatively small, but significant, differences between treatments. There was a high % ground cover with the weed Fathen at G's, and it was possible that this was associated with the high downy mildew at G's. However, there was no relationship between downy mildew and the % ground cover of weed when investigated on a plot by plot basis.

There were significant differences between fungicides in their effectiveness against Botrytis at Merrymac; Botrytis was not observed at G's in any plot, possibly due to cultivar differences. At Merrymac, Switch gave 0% Botrytis, compared to 4% Botrytis in the untreated plots. The other fungicides with the exception of Octave gave 50% control or better; Octave had similar levels of Botrytis to the untreated.

Lettuce weights at harvest were significantly different between treatments at Merrymac, but not at G's, although at both sites, Switch had the lowest weights. At Merrymacs, the lower weights were expected from the plant stunting noted on 25 April, two weeks after the second spray. At G's, no plant stunting was noted, and there was not a significant difference between treatments for the untrimmed or trimmed weights. It must be noted however that Switch is not approved for three sprays on lettuce, and therefore weight results need to be interpreted with caution. Other fungicides such as Amistar (strobilurin) would normally be applied as part of a spray programme with other active ingredients.

Originally, the biocontrol agent Contans was proposed as one of the treatments in the fungicide trial. However, in the initial stages of the project when the location of experiment

sites was negotiated, the fields on offer were on rented land. In practice this meant that the fields were not accessible until immediately prior to planting. This made it difficult to give a fair test for Contans, because it needs to be incorporated into soil at least three months before planting, as well as at time of planting. Therefore, it was decided not to include Contans as a treatment in the fungicide trial in year 1. In discussion with growers at the first project meeting, the decision was made to omit Contans from year 2 of the project because of the problems with use of Contans on vegetable crops and the difficulty of interpreting results from one experimental year.

Contans is intended as a long term treatment for soils, and for optimum results it should be applied to a whole farm and other fields over several years, to reduce disease from airborne spores from adjacent fields. This would need co-operation between farmers. These requirements make it difficult to include Contans as one treatment in a fungicide trial, and it may be only possible to test Contans properly in a dedicated trial. Contans may be better suited for use where minimum tillage is possible, such as oilseed rape. Vegetable crops usually require ploughing or cultivation immediately before planting, which may bring viable sclerotia to the surface, and therefore Contans will need to be applied at each planting. Contans is effective only in the depth to which it is incorporated, so cultivations subsequent to an application should not be deeper than the initial incorporation depth. Therefore it may be difficult to control Sclerotinia with Contans in crops such as carrots which require deep ploughing.

Technology transfer

There have been no technology transfer activities in the first year.

References

- Clarkson JP, Young CS, 2005. 'Forecasting Sclerotinia in field-grown lettuce' Defra project HH3215TFV, Jan 2003 – Dec 2005, ADAS and Warwick HRI.
- Clarkson JP, Phelps K, Whipps, JM, Young CS, Smith JA, Watling M, 2007. Forecasting Sclerotinia disease on lettuce: a predictive model for carpogenic germination of *Sclerotinia sclerotiorum* sclerotia. *Phytopathology* **97**, 621-631.
- Collier, R, 2007. New MORPH takes shape. HDC News, no. 135, July/Aug 2007.
- Rickard PC, 1978. Perforated polythene for forwarding vegetable crops. ADAS Horticulture, Cambridge.

- Schafer MR, Kohn LM, 2006. An optimized method for mycelial compatibility testing in *Sclerotinia sclerotiorum. Mycologia* **98**, 593-597.
- Twengstrom E, Sigwald R, Svensson C, Yuen J. 1998. Forecasting Sclerotinia stem rot in spring sown oilseed rape. *Crop Protection* **17**, 405-411.
- Young CS, Clarkson JP, Smith JA, Watling M, Phelps K, Whipps JM, 2004. Environmental conditions influencing *Sclerotinia sclerotiorum* infection and disease development in lettuce. *Plant Pathology* **53**, 307-397.

Appendix 1

Applications to commercial crops at G's and Merrymac sites, 2007

Site 1: Merrymac Salads, applications made to field CW 63, lettuce crop April 2007 Harvest Date Product Interval Ha Rate/Ha Units GS Water Vol MAPP / Active Ingredient 18/02/2007 Touchdown Quatt 0 12.17 2.500 lt 100 10608 Glyphosate Problem Volunteers/Broad Leaved Weeds Comment authorised by john hall JH01 glyphosate - authorised by john hall JH01 glyphosate Field Comment completed Justification Broad Leaved Weeds/Volunteers Start 9.30 Finish 10.45 Wind NE 5 Temp Code 2 6-10 C Spray Quality Medium Operator Bryan Porter 21/02/2007 Ammonium Nitrate 12.10 182.000 Kg 0 0 Comment authorised by john hall JH 002 Start 15.30 Finish 16.00 Wind SW 4 Temp Code 0-5 С 1 Operator Johnny Clarke 21/02/2007 9-9-22.5 0 0 12.17 560.000 Kg Comment authorised by john hall JH 003 Finish 18.50 Wind SW 3 6-10 С Start 17.40 Temp Code 2 Nick Smith Operator 0 12025 22/02/2007 CleanCrop Hyde 0 12.17 4.000 Kg Metaldehyde Comment -Justification Slugs NE 5 Temp Code С Start 10.00 Finish 10.45 Wind 6-10 2 Operator Bryan Porter 15/03/2007 Lettuce Starter 12.17 210.000 Lt 0 24/03/2007 Rovral WP 7 11.75 400 11694 0.119 kg Iprodione Problem Botrytis/Manganese deficiency authorised by john hall - JH014 - JH014 Comment Field Comment completed

Justification Botrytis Start 9.30 Finish 11.00 Wind Operator Bryan Porter	SW 10 Temp Code 3 11-15 C	Spray Quality Medium
24/03/2007 Manganese Sulph Problem Botrytis/Manganese deficience Comment authorised by john hall - JH0 ⁻		400
Field Comment completed Start 9.30 Finish 11.00 Wind Operator Bryan Porter	SW 10 Temp Code 3 11-15 C	Spray Quality Medium
27/03/2007 Alpha Propachlo Propachlor Problem Broad Leaved Weeds Comment authorised by john hall - JH0 Field Comment completed Justification Broad Leaved Weeds	0 11.50 3.000 lt 16 - JH016 - JH016	0 4873
Start 15.30 Finish 17.12 Wind Operator Bryan Porter	Ea 5 Temp Code 3	11-15 C
02/04/2007 Alpha Propachlo Propachlor Problem Broad Leaved Weeds Comment authorised by john hall - JH07 Field Comment completed Justification Broad Leaved Weeds Start 10.20 Finish 11.32 Wind	0 11.50 2.000 lt 15 - JH015 - JH015 NE 10 Temp Code 3 11-15 C	400 4873 Spray Quality Medium
Operator Bryan Porter		
02/04/2007 Comrade Chlorpropham Problem Broad Leaved Weeds Comment authorised by john hall - JH07 Field Comment completed Justification Broad Leaved Weeds Start 10.20 Finish 11.32 Wind	0 11.50 2.000 lt 15 - JH015 - JH015 NE 10 Temp Code 3 11-15 C	400 10181 Spray Quality Medium
Operator Bryan Porter		
06/04/2007 Signum Boscalid+Pyraclostrobin Problem Botrytis/Manganese deficience Comment authorised by john hall - JH02 Field Comment completed Justification Botrytis Start 15.30 Finish 16.41 Wind		400 11450 Spray Quality Medium
Operator Bryan Porter		opray during mouldin

06/04/2007 Manganese Sulph Problem Botrytis/Manganese deficience Comment authorised by john hall - JH02 Field Comment completed Justification Manganese deficiency Start 15.30 Finish 16.41 Wind		-	400 Spray Quality Medium
Operator Bryan Porter			
06/04/2007 Bittersaltz	0	11.50 2.000 Kg	400
Problem Botrytis/Manganese deficience Comment authorised by john hall - JH02 Field Comment completed Justification Magnesium deficiency		•	
Start 15.30 Finish 16.41 Wind Operator Bryan Porter	NE 5	Temp Code 3 11-15 C	Spray Quality Medium
10/04/2007 Manganese Sulph Problem Manganese deficiency Comment authorised by john hall - JH03 Field Comment completed	0 33	11.50 2.000 Kg	400
Justification Manganese deficiency Start 17.00 Finish 18.36 Wind Operator Bryan Porter	NW 5	Temp Code 3 11-15 C	Spray Quality Medium
16/04/2007 Fubol Gold WG	14	11.50 1.900 kg	400 10184
Mancozeb+Metalaxyl-M Problem Mildew/Magnesium deficienc Comment authorised by john hall JH037 Field Comment completed Justification Mildew			
Start 17.35 Finish 19.00 Wind Operator Bryan Porter	NW 5	Temp Code 4 16-20 C	Spray Quality Medium
16/04/2007 Manganese Sulph Problem Mildew/Magnesium deficienc Comment authorised by john hall JH037 Field Comment completed			400
Justification Manganese deficiency Start 17.35 Finish 19.00 Wind Operator Bryan Porter	NW 5	Temp Code 4 16-20 C	Spray Quality Medium
16/04/2007 Bittersaltz Problem Mildew/Magnesium deficience Comment authorised by john hall JH037 Field Comment completed Justification Magnesium deficiency		-	400

Start 17.35 Finish 19.00 Wind Operator Bryan Porter	NW 5	Temp Code 4 16-20 C	Spray Quality Medium	
18/04/2007 Manganese Sulph Problem Manganese deficiency Comment authorised by john hall JH034 Field Comment completed Justification Manganese deficiency	0 3 - JH038	11.50 2.000 Kg - JH038 - JH038 - JH038	400	
Start 10.00 Finish 11.30 Wind Operator Bryan Porter	NW 5	Temp Code 4 16-20 C	Spray Quality Medium	
27/04/2007 Amistar Azoxystrobin Problem Mildew/Manganese deficience Comment authorised by john hall JH049 Field Comment completed Justification Mildew			400 10443	
Start 13.30 Finish 14.48 Wind Operator Bryan Porter	NE 10	Temp Code 4 16-20 C	Spray Quality Medium	
27/04/2007 Karamate Dry FI Mancozeb Problem Mildew/Manganese deficience Comment authorised by john hall JH049 Field Comment completed Justification Mildew			400 12691	
Start 13.30 Finish 14.48 Wind Operator Bryan Porter	NE 10	Temp Code 4 16-20 C	Spray Quality Medium	
27/04/2007 Decis Deltamethrin Problem Mildew/Manganese deficience Comment authorised by john hall JH049 Field Comment completed Justification Caterpillar			400 7172	
Start 13.30 Finish 14.48 Wind Operator Bryan Porter	NE 10	Temp Code 4 16-20 C	Spray Quality Medium	
27/04/2007 Manganese Sulph 0 11.50 2.000 Kg 400 Problem Mildew/Manganese deficiency/Magnesium deficiency/Caterpillar Comment authorised by john hall JH049 - JH049 - JH049 - JH049 Field Comment completed Justification Manganese deficiency				
Start 13.30 Finish 14.48 Wind Operator Bryan Porter	NE 10	Temp Code 4 16-20 C	Spray Quality Medium	
27/04/2007 Bittersaltz	0	11.50 2.000 Kg	400	

Comment authorise Field Comment comp Justification Magnesi Start 13.30 Finis	um deficiency	, ,		049	Spray Quality	Medium	
10/05/2007 Farm	-Fos	0	11.50	2.000 Lt		400	
Comment authorise Field Comment icb 0 Justification Multi ele Start 8.00 Finisi	ed by john hall JH06 26 029 028 sprayed ment deficiency	0 - JH060			ficiency/Aphids Spray Quality	Medium	
	anese Sulph	0	11.50	2.000 Kg		400	
	ed by john hall JH06 26 029 028 sprayed	0 - JH060	-	ciency/Magnesium de ted	ficiency/Aphids		
Start 8.00 Finis Operator Bryan	n 9.30 Wind n Porter	We 8	Temp C	ode 4 16-20 C	Spray Quality	Medium	
10/05/2007 Bitter	saltz	0	11.50	2.000 Kg		400	
Comment authorise Field Comment icb 0 Justification Magnesi Start 8.00 Finisi	ed by john hall JH06 26 029 028 sprayed um deficiency	0 - JH060	-		ficiency/Aphids Spray Quality	Medium	
10/05/2007 Topp	el 10	0	11.50	147.436 ml		400	8772
Cypermethrin Problem Caterpillar/Multi element deficiency/Manganese deficiency/Magnesium deficiency/Aphids Comment authorised by john hall JH060 - JH060 Field Comment icb 026 029 028 sprayed on 6/5/07 now completed Justification Caterpillar Start 8.00 Finish 9.30 Wind We 8 Temp Code 4 16-20 C Spray Quality Medium Operator Bryan Porter Image: Spray Porter Spray Porter Spray Porter							
10/05/2007 Aphc Pirimicarb	x	3	11.50	300.000 g		400	10515
Problem Caterpillar/Multi element deficiency/Manganese deficiency/Magnesium deficiency/Aphids Comment authorised by john hall JH060 - JH060 Field Comment icb 026 029 028 sprayed on 6/5/07 now completed							
Justification Aphids Start 8.00 Finis		We 8	Temp C		Spray Quality	Medium	

Safe Harvest Date -13/05/2007

Site 2: G's site, applications made to Buckinghams Field, lettuce crop April 2007

J B Shropshire &Sons Ltd (Engine Farm), JBS&S Ltd Engine Farm (WLF Operations) Field: Buckinghams Field Area: 20.00 ha Projected Harvest Date: 02/05/2007 Crop: Iceberg Lettuce Drilled Area: 15.46 ha (01/09/06 to OZ/05/07 Earliest Harvest Date: 02/05/2007 1222

Application date, No, Operator, Area, Product, Active, Reason, Rate, Water Vol., (SOLA no), HI Days

19/02/07 609 Jamie Smith 18.00 N37, 135.00 L/ha 17/02/07 629 omex 20.00 7-18-7 700.00 kgslha 20/02/07 659 15.46 Hydro Starter Flow 184.00 L/ha

07/03/07 2,374 Jamie Smith 15.46 ROVRAL WP Iprodione, Botrytis, 0.20 kg/ha/ha, 400 Lts/ha, 7

07/03/07 2,374 Jamie Smith 15.46, Manganese Sulphate, Trace Element 2.00 kgs/ha/ha, 400 Lts/lha

07/03/07 2,374 Jamie Smith 15.46 BTTTERSALTZ Trace Element 2.00 kgs/ha/ha 400 Lts/lha

07/03/07 2,374 Jamie Smith 15.46 CROPUFT Potash Nitrogen Phosphate Magnesium kgs/ha/ha Trace Element 0.60 400 Lts/lha

08/03/07 2.375 Jamie Smith 15.46 RAMROD FLOWABLE Propachlor BL Weeds 3.00 lts/ha/ha 450 Lts/ha (SOLA 1159/02) 42

23/03/07 2,377 M Sporle 15.46 SLUGGO Ferric Phosphate Slugs 11.32 kgs/ha/ha

26/03/07 2,380 Jamie Smith 15.46 COMRADE Chlorpropham BL Weeds 2.00 Lts/ha/ha 500 Lts/ha

28/03/07 2,391 Jamie Smith 15.46 Signum Boscalid Pyraclostrobin Botrytis 1.50 Unit/ha/ha 400 Lts/ha 14

28/03/07 2,391 Jamie Smith 15.46 Manganese Sulphate Trace Element 2.00 kgs/ha/ha 400 Lts/ha

28/03/07 2,391 Jamie Smith 15.46 CROPUFT Potash Nitrogen Phosphate Magnesium Trace Element 0.60 kgs/ha/ha 400 Lts/ha

11/04/07 2,430 Jamie Smith 15.46 INVADER DImethomorph Mancozeb Mildew control 2.00 kgs/ha/ha 400 Lts/ha (SOLA 3044106) 21

11/04/07 2,430 Jamie Smith 15.46 Manganese Sulphate Tace Element 2.00 kgs/ha/ha 400 Lts/ha

11/04/07 2,430 Jamie Smith 15.46 CROPUFT Potash Nitrogen Phosphate Magnesium kgs/ha/ha Trace Element 0.60 400 Lts/ha

18/04/07 2,443 Jamie Smith 15.46 FUBOL GOLD Mancozeb Metalaxyl-m Mildew control 1.90 kgs/ha/ha 400 Lts/ha (SOLA 2141/03) 14

18/04/07 2,443 Jamie Smith 15.46 BITERSALTZ Trace Element 2.00 kgs/ha/ha 400 Lts/ha

18/04/07 2,443 Jamie Smith 15.46 Manganese Sulphate Trace Element 2.00 kgs/ha/ha 400 Lts/ha

Appendix 2

Sclerotial germination model: replacing soil water potential threshold with a threshold triggered by a temperature-rainfall function

The new rainfall-temperature based function and original water potential function both determine whether germination can progress. By assigning a 1 when the function indicates the soil is moist enough and zero otherwise, Figure 1a and 1b compare the behavior of each function at each time step for the Cheshire and Norfolk sites in 1994 in order to assess the fit of the new function. Note that the horizontal portions of the traces are associated with dry weather when the germination cannot proceed and in general the two functions flatten off at the same time. It is important to note that in the original model the water potential for Cheshire 2004 was set at -8 kPa and the water potential at the Norfolk site in 2004 at -4 kPa, whereas the new temperature rainfall function is generic and therefore applicable to both sites and requires no local calibration. A comparison of the two models in predicting the T50 is presented later in this appendix.

Fig 1(a).

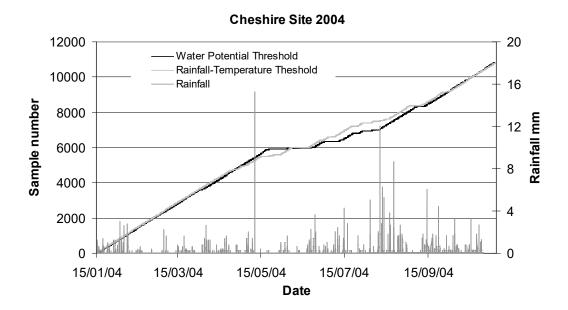


Figure A1(b).

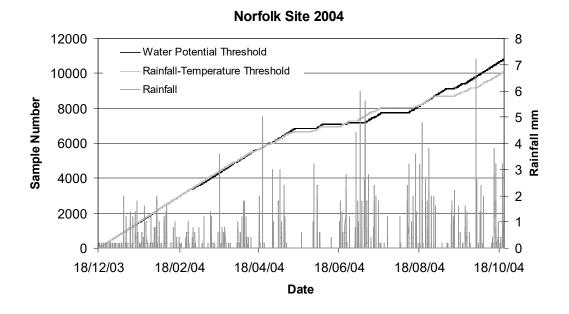


Figure. A1 (a) and (b). A comparison of the water potential threshold of the original model and the new temperature-rainfall function developed for the forecasting model.

Model validation

New model predictions of T10 with the new rainfall-temperature function and the modification to predict T10 directly have been compared to the results published in Clarkson et. al., (2006) for burial dates at each site where data was available. The results for the original model include the original water potential function which uses its optimised value for each site and in each year. In addition the T10 in the original model is calculated by first estimating T50, then applying the log-normal curve and reading off the time to 10% germination (T10). For both the original model and the new model predictions the parameters for the local isolates have been used which is isolate TM at Norfolk and isolate 13 at Cheshire. Table A1 below presents some simple comparative statistics (mean difference between observed and predicted T10's (days); the standard deviation; the largest difference between observed and modelled across the burials; the standard error on the estimate of the mean difference and the standard error on the standard deviation) across all the data for example the average difference in days between the observed and predicted T10 at the Cheshire site was -3.8 days for the original model and 4.4 days for the new model.

Figs A3, A4 and A5 compare the original models estimate of T10 to the new models estimate. The new model performs better at the Cheshire site compared to the original model even though it calibrated of the water potential each year. For both sites the new model is performing at a similar level overall but has the major advantage of the inclusion of the generic rainfall-temperature function which requires no site local calibration and the method of estimating T10 does not require the T50 to be calculated first.

Table A1. Comparative statistics (in days) for T10's estimated using the model of Clarkson et al (2006) and the new meta-model.

Site	Cheshire	
	••••••	
Method	Original Model	Meta-Model
Avg. difference Obs-Pred	-3.8	-4.4
Std Deviation Obs-Pred	22.3	16.0
Largest difference Obs-Pred	-56.7	-38.6
Number of Samples	24.0	24.0
Standard error mean	-0.8	-0.9
std er stdev	3.2	2.3
	Norfolk	
	Original Model	Meta-Model
Avg. difference Obs-Pred	-4.9	-1.4
Std Deviation Obs-Pred	16.6	18.9
Largest difference Obs-Pred	-31.0	-48.4
Number of Samples	22.0	22.0
std er mean	-1.0	-0.3
std er stdev	2.5	2.8
	Both Sites	
	Original Model	Meta-Model
Avg. difference Obs-Pred	-4.3	-3.0
Std Deviation Obs-Pred	19.6	17.3
Largest difference Obs-Pred	-56.7	-48.4
Number of Samples	46.0	46.0
std er mean	-0.6	-0.4
std er stdev	2.0	1.8

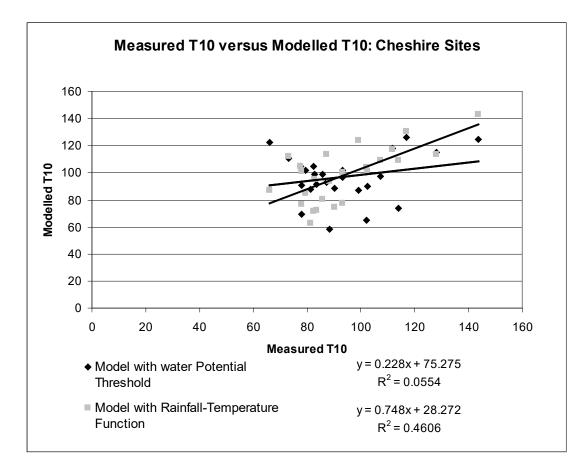


Figure A3. Original modelled T10's for Cheshire burials 2000-2005 versus new modelled T10's.

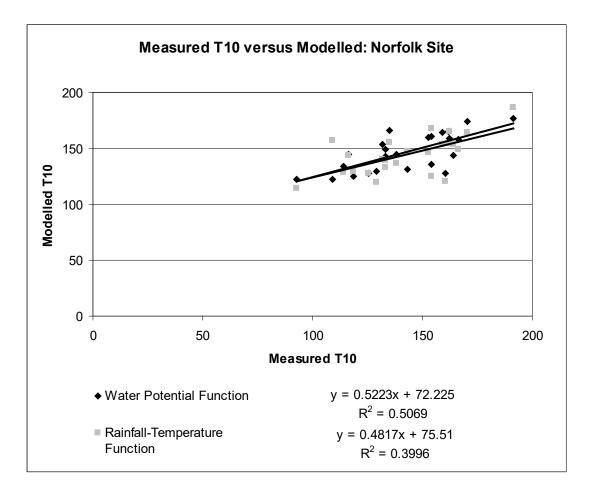


Figure A4. Original modelled T10's for Norfolk burials 2000-2005 versus new modelled T10's.

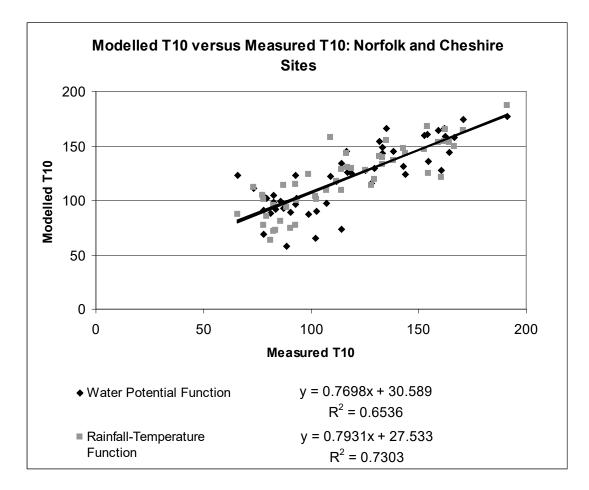


Figure A5. Original modelled T10's for Cheshire and Norfolk burials 2000-2005 versus new modelled T10's.