Project title: Narcissus: white mould decision support system

Project number: BOF 56

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Location: Commercial crops of Narcissus in West Cornwall

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Key words: Narcissus, white mould, disease control, *Ramularia vallisumbrosae, BOF 56*

Contents

Grower Summary

Headline

- The application of an accurate fungicide programme for narcissus will minimise the development of the leaf disease white mould and reduce the number of sprays required.
- A reduced number of fungicides will mean that applications are made in an environmentally friendly, sustainable and cost effective way.

Background and expected deliverables

The management of narcissus leaf diseases is of major concern to bulb growers in Cornwall and the Isles of Scilly. Producing disease free bulbs and foliage is important in meeting the high quality specifications demanded by the export trade and by the multiple retail sector. In addition, pest and disease management in crops is expected to be achieved with minimal, and justified, use of pesticides.

In the past decade, the bulb growers have become more concerned with the large increase in the incidence of fungal foliar diseases, particularly ‘white mould’. The symptoms of white mould can be very dramatic, causing early senescence, and is estimated by growers to exact a loss in yield of about 10% annually; losses could be much greater in specific situations. For many years the control of daffodil foliar diseases has been by a more or less non-specific programme of fungicide sprays. Up to seven sprays, often mixtures of different active ingredients may be used on these crops each year.

A Defra and HDC funded ‘Horticulture LINK’ programme completed in 2002 studied the factors leading to infection of daffodil leaves with the white mould fungus. This showed that infection was dependent on temperature and leaf wetness duration, and a mathematical model was formulated to describe this relationship. The model allows infective periods to be identified through interrogating data from temperature and surface wetness sensors sited in the crop, enabling growers to target fungicide applications to the key periods when they will be most effective. Field trials, part of the same project, showed that the number of fungicide applications used on a daffodil crop might be reduced by half, without loss of control, through such targeting, ensuring that fungicide applications are made in an environmentally friendly way. The present project aims to translate the research findings into a practical system that growers and advisors can use, by validating the model and delivering it as a user-friendly, practical ‘spray timing system’.

The expected deliverables from this project are:

- Clear guidelines on application rates, timing and number of fungicide treatments needed to control white mould.

- An evaluation of spray timing to establish a decision support system based on information provided from the ‘Horticulture LINK’ programme.

Summary of the project and main conclusions

- Critical weather conditions favouring white mould development were temperatures between 5 and 10°C combined with leaf wetness durations of 12 to 24 hours.
- At Rosewarne the highest prediction infection scores occurred on 18 January. At Nanceddan the highest score occurred on 7 April, coinciding with an expected infective period.
- Disease forecasting models can provide information on the optimal timing of fungicide application for controlling crop disease.
- By developing specific models which summarise important life-cycle stages, and using in-field weather data, information can be obtained to show when conditions are critical for pathogen development in crops.
- By monitoring the environmental conditions necessary for infection, the infection risk can be determined.

Science Section

Introduction

Daffodil (narcissus) growing is an important component of the agriculture and horticulture industry in Cornwall and the Isles of Scilly. Bulb crops have been grown here comercially since the late-19th century, becoming a part of the landscape as well as a source of employment, income and enjoyment. Since the 1970s, entrepreneurial growers have developed a healthy export trade in both daffodil bulbs and cut flowers. UK daffodil bulbs and flowers are held in high esteem and they are exported all over Europe and North America.

The control of fungal diseases is of major concern to bulb growers, since the production of disease- and pest-free bulbs is vital in meeting the exacting specifications of the export trade and multiple-retail sector. It is expected that pest and disease management be achieved with only minimal use of pesticides, and should be justified on a case-by-case basis. In the past decade, UK bulb growers in Cornwall have become concerned with epidemics of the fungal foliar disease white mould caused by *Ramularia vallisumbrosae*.

For many years the control of daffodil foliar diseases has been by a more or less non-specific programme of fungicide sprays. Up to seven sprays, often mixtures of different active ingredients, may be used on these crops each year. As little specific information is known about the control of white mould, fungicides have been applied more in the hope of a general fungistatic efficacy than based on any assurance as to their fungicidal effectiveness. This changed following a research project carried out in 1998 to 2002 under the 'Horticulture LINK' programme, and funded by Defra, the Horticultural Development Council (HDC), nine bulb growing companies (considered to represent 70% of the UK daffodil acreage) and Aardware Design, a producer of environmental sensing devices.

The 'LINK' project included experiments in controlled environment cabinets to study the factors leading to infection of daffodil leaves with the white mould fungus. These studies demonstrated that infection was dependent on temperature and leaf wetness duration, and mathematical relationships (models) were formulated to describe this relationship. These mathematical models allow infective periods to be identified through interrogating data from temperature and surface wetness sensors sited in the crop, enabling growers to target fungicide applications to these key periods when they will be most effective. Field trials, part of the same project, showed that the number of fungicide applications used on a daffodil crop might be reduced by half, without loss of control, through such targeting, ensuring that fungicide applications are made in an environmentally friendly and sustainable, and also highly effective and specific, way. This project aims to translate the research findings into a practical system that growers and advisors can use, by:

- Validating (i.e. testing and refining if necessary) the model to make sure it works in practice);
- Delivering the validated model as a user-friendly, practical 'spray timing system' for bulb growers, informing them when it is best to apply fungicides (and, just as importantly, when it is unnecessary or inappropriate to do so).

The 'LINK' project was subjected to economic scrutiny by independent assessors, as part of a quinquennial review of the HDC. This assessment showed the project would have a very high economic benefit if carried through to the development phase that enabled its findings to be applied by the bulb industry.

Materials and Methods

Sites for crop monitoring

In autumn 2004, two second-year, Cornish daffodil crops were selected for monitoring (see Table 1). As far as practical, the selected crops were typical of the region. In each crop an area *ca.* 0.2ha in extent was clearly marked with corner posts and other markers, and it was agreed with the owner that no fungicide sprays would be applied through the second year of the crop within this designated area. In all other respects, it was agreed that the crop would be farmed according to the grower's normal commercial practices. The central 0.1ha of the designated area was demarcated for monitoring and observation, leaving the surrounding area as a buffer zone for protection from any spray drift from adjacent crops.

Table 1. Cornish white mould monitoring sites.

Year monitored	*Owner's name*	*Site name*	*Grid reference*	*Cultivar, year*
2004-2005	Rosewarne Rosewarne Camborne TR14 0AB	Rosewarne, Camborne	SW 6441	'Carlton' 2nd
2004-2005	Winchester Growers Ltd Varfell Farm Long Rock Penzance TR20 8AQ	Nanceddan, Ludgvan, Penzance	SW 5034	'Planet' 2nd

Weather data

A meteorological data logger ('Smaartlog'; Intelligent Micro Design Ltd.) was set up close to the centre of each monitoring area prior to crop emergence. The loggers, powered by battery and solar panel and downloadable *via* a modem and digital cell telephone, were provided with sensors recording soil and air temperature, relative humidity, surface wetness and rainfall, at 30-minute intervals.

Trap plant production

Bulbs of daffodil 'Carlton', grade 12-14cm circumference, were allocated in August 2004 from a stock grown at Warwick HRI, Kirton, Lincolnshire. For comparability with the commercial, second-year daffodil crops being monitored, the bulbs were not given hot-water treatment nor any fungicide applications from lifting onwards. The bulbs were stored at 17°C until early-October 2004, when they were planted in a standard fashion, five bulbs per 20cm-diameter, 4L-capacity plant-pot using a peat/sand/John Innes mix. The pots were stood outdoors and covered with fleece to protect the plants from extreme weather, and were kept well watered. In December, the plant-pots were transported to Camborne, Cornwall and stood outdoors until required. Before exposure (see below), the foliage on half the allocated pots was wounded in a standard fashion by brushing with a stiff brush, the other half remaining undamaged as controls.

Disease symptoms

White mould leaf lesions appeared in spring, often singly, in the upper one-third of the leaf, and near the mid-line. The lesions were elongated areas often 5 – 10 mm in length, with the degraded leaf surface presenting as sunken grey-green to yellowish areas. When sporulating, the lesion surface appeared powdery and characteristically creamy-white in colour. Later, rows of minute black sclerotia-like bodies (visible with a hand-lens) were present in the lesions. Sometimes the affected area became degraded, leaving a ragged hole in the leaf. In serious cases further lesions appeared elsewhere on the leaves and flower stalk, becoming elongated and coalescing. White mould often occurred in prominent patches of the crop 1 – 2 m across. As the disease progressed, the leaves died-back from the tip, becoming dry and brown, sometimes in a matter of days. Instances were seen where the flower stalks were similarly affected, although in other cases they did not appear to be attacked, remaining erect among the dead foliage. No symptoms were seen on flowers.

Crop and disease monitoring and spore trapping

The selected areas of crops were checked at weekly intervals from December onwards, and the crop stage and the date of first appearance of white mould symptoms was recorded. Following the appearance of first symptoms, disease levels were assessed weekly for the incidence and severity of disease. The central, 0.1ha area was walked in an X-pattern starting from a marked corner; on crossing ridges a 0.5m-long section (sub-sample) was delimited (with a ruler) at the intercept, with 50 sub-samples were scored at each assessment. The incidence and severity of white mould were scored in each of the 50 sub-samples according to the scale shown in Table 2. Overall incidence and severity scores were calculated by summing the scores for all 50 sub-samples. The crop growth stage and percentage of foliage senescent/dead was also noted.

Table 2. White mould incidence and severity scales

Score	*Incidence per 0.5m plot*	*Score*	*Severity within each 0.5m plot*
0	None	0	None
1	1 or 2 leaves affected	1	Single lesions
2	>2 but <10 leaves affected	2	Single lesions or occasionally >1 lesion per leaf
3	>10 leaves but <50% leaves affected	3	Generally 2 or more lesions per leaf
4	>50% but <100% leaves affected	4	Lesions coalescing to form larger damaged areas
5	All leaves affected	-	-

Following shoot emergence and continuing until the end of the growing season, a spore trap was set up near the centre of each area. At weekly intervals the recording tape of the spore trap was replaced, the exposed tape being refrigerated and sent to Warwick HRI, Wellesbourne, University of Warwick, for examination.

Also following the appearance of first symptoms, pot-grown daffodil ‘Carlton’ trap plants were placed close to crop foliage near the centre of each area for exposure periods of 24h. The plants were put out on Monday through Thursday and collected Tuesday through Friday. For each period, six plant-pots – three with undamaged plants (controls) and three with damaged leaves (a stiff bristle nail-brush was drawn across the leaves in a standard fashion) – were used. Following collection from the field sites, the exposed plant-pots were placed in a frost-protected (minimum maintained temperature, 3ºC) well ventilated (10ºC) glasshouse free of other potentially infective plant material, at Duchy College, Rosewarne, Camborne, Cornwall. Further non-exposed control pots were moved straight to the glasshouse without exposure (three pots per week). The three replicate plant-pots in each set were arranged in the glasshouse in three blocks, keeping all pots well spaced from one another. The plant-pots were kept well watered during this time, using bottom-watering into saucers to avoid spreading infection. Plants were examined for disease lesions at weekly intervals, and once symptoms were present the number and extent of lesions in each pot was recorded at 2-weekly intervals over a period of 14 weeks.

Results

Meteorological data

Temperature, relative humidity, rainfall and leaf wetness data are shown for Rosewarne and Nanceddan in Figures 1-4. Previous research (Hanks *et al.*, 2003) concluded that the critical weather conditions favouring white mould infection were temperatures between 5 and 10ºC combined with leaf wetness durations of 12 to 24 hours. The weather data obtained can therefore be analysed to determine likely infective periods. Unfortunately some logger malfunctions and battery loss occurred and some data were lost, always a potential problem at a remote site; we believe these problems have been rectified and three meteorological stations will be deployed in 2006 to compensate for the previous year's loss.

During the period studied, at Rosewarne average air temperatures fell generally into the 5-10ºC band over 17-19 January and 28 January to 5 February, while there were periods of leaf wetness >12 hours on 17 and 19-23 January and on 1-3 and 11 February. These periods corresponded to periods of relative humidity >95% and to days with heavy rainfall. The data suggest that 17 and 19 January and 1-3 February would be the key critical periods for white mould infection at the Rosewarne site.

Carrying out a similar analysis of the data for Nanceddan, the likely infective periods indicated were 11-13 March, 16-19 March, 28-29 March, 3-7 April and 12-17 April.

Predicted infection score

The white mould infection model was run with the above temperature and leaf wetness data for the two sites, and the predicted scores are presented in Figures 5-6. At Rosewarne the highest predicted infection score occurred on 18 January, corresponding with an infection period derived empiracly from the meteorological data (see above). At Nanceddan, the highest score occurred on 7 April, again coinciding with an expected infective period.

Crop and disease monitoring

The numbers of plots showing white mould symptoms are shown in Figures 7-8. At Rosewarne, white mould symptoms were first seen in early-March (14 plots affected), some 6 weeks after the highest predictive score was recorded, and numbers increased to >40 over the next three weeks. At Nanceddan, white mould symptoms were first seen at the beginning of March (19 plots), steadily increasing over the growing season with all plots showing symptoms by late-April. Here, though predictive scores for the early part of the year are not available, the frequently high predictive scores seen over the late-March to mid-April may be related to the very high disease incidence and severity seen at the site.

The incidence and severity scores at Nanceddan increased steadily from the beginning of March, while at Rosewarne scores increased rapidly from mid-March and peaked at the end of March (Figures 7-8).

Trap plants

White mould lesions were seen on only three of the trap plants from Nanceddan, on plants exposed starting 12 March (on one undamaged and one damaged plant) and 13 April (on one damaged plant). No lesions were seen on trap-plants from Rosewarne.

Spore traps

Only two dates resulted in trap plant infection and at Nanceddan only, spore tapes from this site for 12 March and 13 April 2005 were examined for the presence of *Ramularia* conidia. Long, thin *Cercosporella*-type spores, possibly scolecospores of *R. vallisumbrosae*, were observed on spore tapes from both these dates, suggesting *R. vallisumbrosae* spores were present in the air during these two dates when trap plant infection occurred. At present there is no additional method of confirming the identity of these structures on the spore tape, or determining if phragmospores of *R. vallisumbrosae* were also present.

Discussion

Disease forecasting models can provide information on the optimal timing of fungicide application for controlling crop disease. By developing specific models which summarise important life-cycle stages, and using in-field weather data, information can be obtained to show when conditions are critical for pathogen development in crops. By monitoring the environmental conditions necessary for infection, the infection risk can be determined. Mathematical relationships (models) describing the effect of temperature and wetness on *R. vallisumbrosae* infection were developed at Warwick HRI in a project funded under the Horticulture LINK programme, and it is envisaged that these will form the basis of a white mould forecasting or spray timing system.

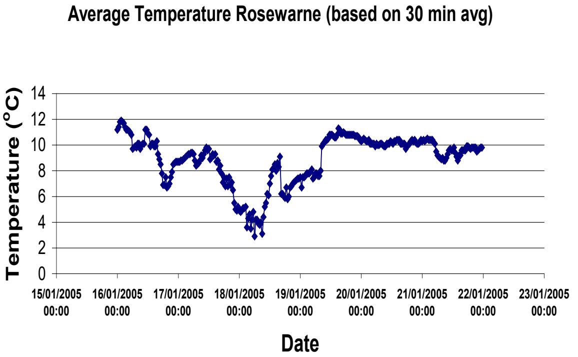
Despite the white mould levels that occurred on the monitored field crops, in the first year of the project white mould lesions were only infrequently seen to develop on trap plants and only limited numbers of *R. vallisumbrosae*-like spores (scolecospores) were found on spore traps, though these corresponding with when the infection of trap plants occurred. If scolecospores are required for infection, these large spores would require specific conditions for their transmission from the crop onto trap and crop plants. It is likely that there are discrete periods for scolecospores production within the crop. Temperatures of 5–10ºC, in conjunction with long periods of high humidity (>95 % relative humidity) are required for the production of scolecospores. Scolecospores were found on spore tapes from both days when trap plant infection was observed, and they appeared not to be present at other times. Conditions conducive to scolecospore production were observed prior to each trap plant period when white mould infection was observed. Additionally, on one occasion (13 April), conditions favourable for scolecospores transmission were also observed. These results suggest that the presence of the scolecospore spore type maybe necessary for the spread of white mould infection. To facilitate the identification of *R. vallisumbrosae* spores, monoclonal antibodies are being developed.

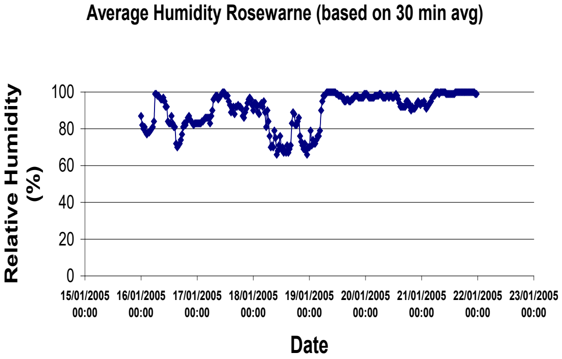
Existing infection models were based on phragmospore inoculation and infection, and indicated that infective conditions for these spores are present on most occasions within the crop, though this could not be extrapolated for scolecospore infection. Further data-sets will be obtained in 2006 to determine if trap plant infection occurs only when scolecospore production takes place. It may be possible to use the existing *Ramularia* infection model to indicate days when scolecospore infection is likely.

References

Hanks, G.R., Kennedy, K. & O'Neill, T.M. (2003). *Narcissus leaf diseases: Forecasting and control of white mould and smoulder.*
Final Report on Horticulture LINK Programme project HORT188, Defra, London, UK (available at http://www2.defra.gov.uk/research/project_data/Default.asp).

Figure 1. Temperature and humidity at Rosewarne. Data averaged over 30-minute periods.


Average Temperature Rosewarne (based on 30 min avg)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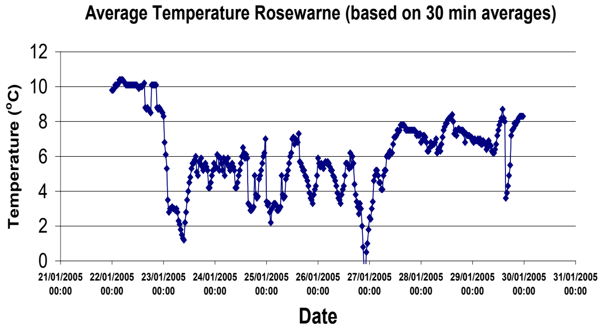
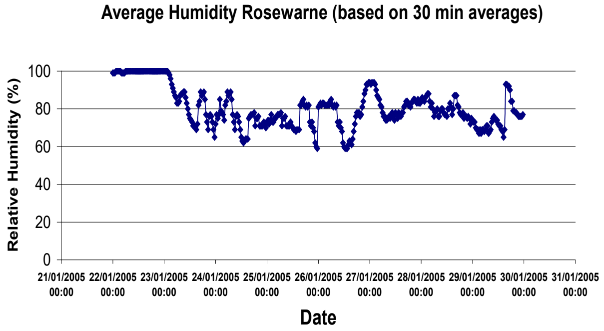

Average Temperature Rosewarne (based on 30 min averages)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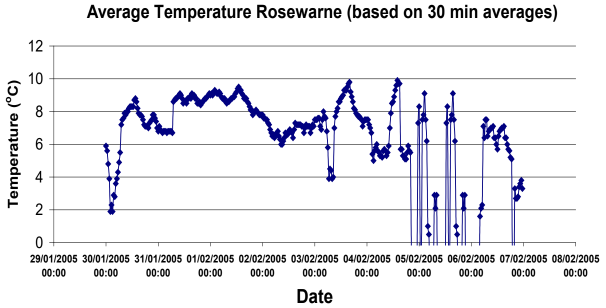
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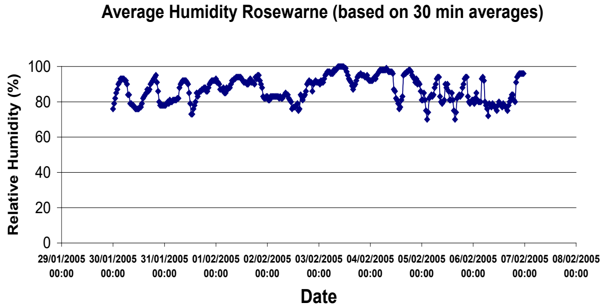

Average Humidity Rosewarne (based on 30 min averages)

Figure 2. Rainfall (▲) and leaf wetness (♦) at Rosewarne. Data averaged over 24-hour periods.

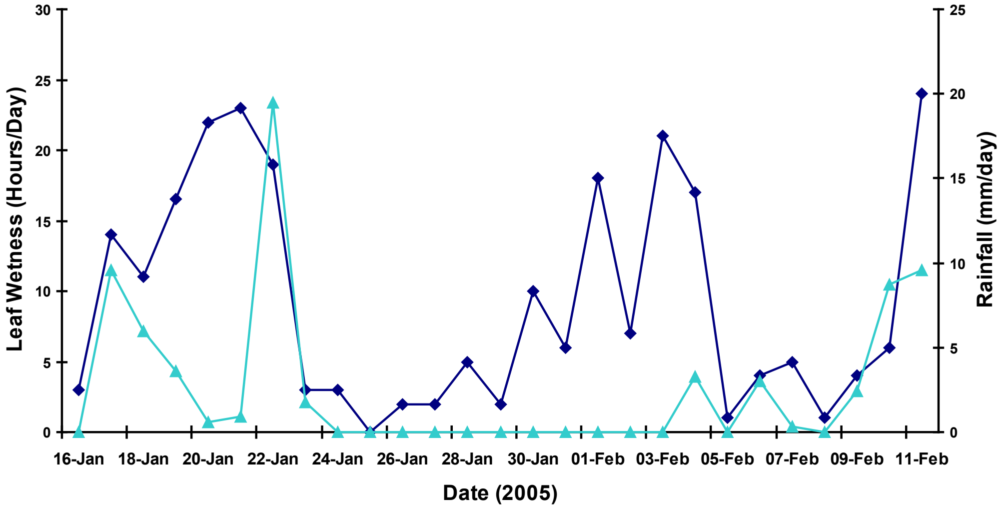
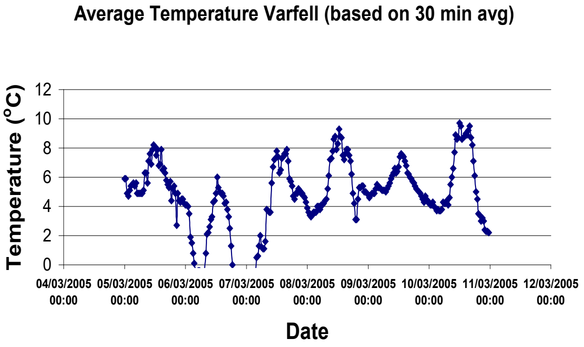
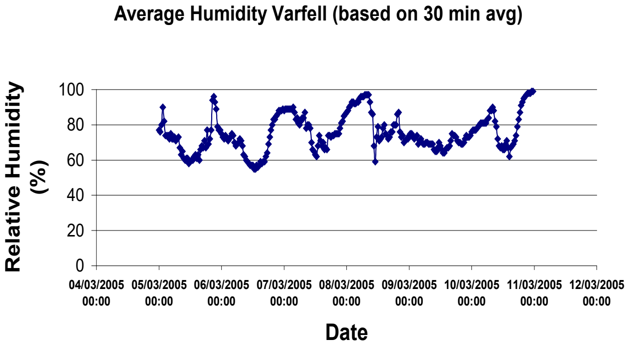
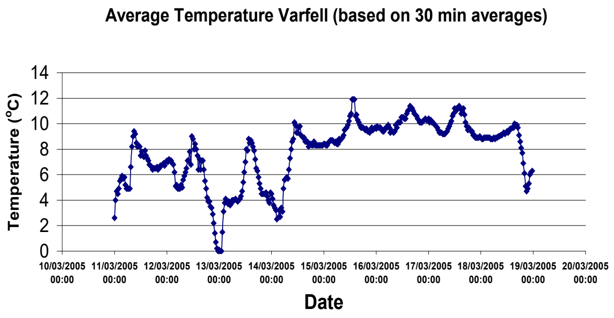
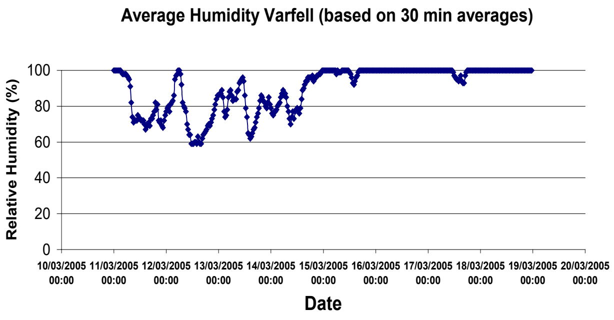


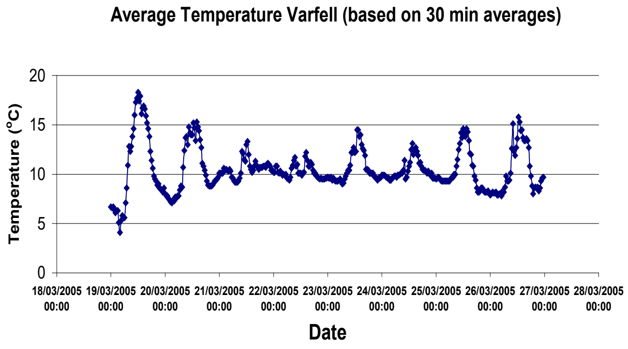
Figure 3. Temperature and humidity at Nanceddan. Data averaged over 30-minute periods.

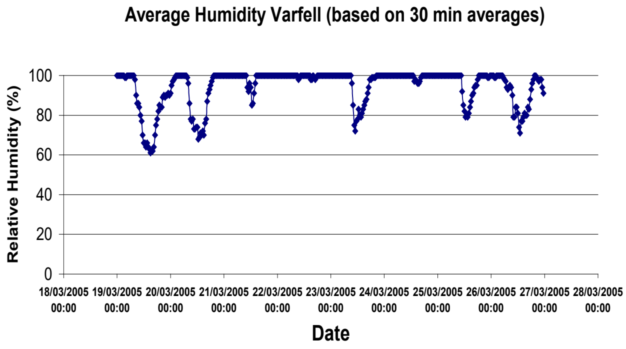


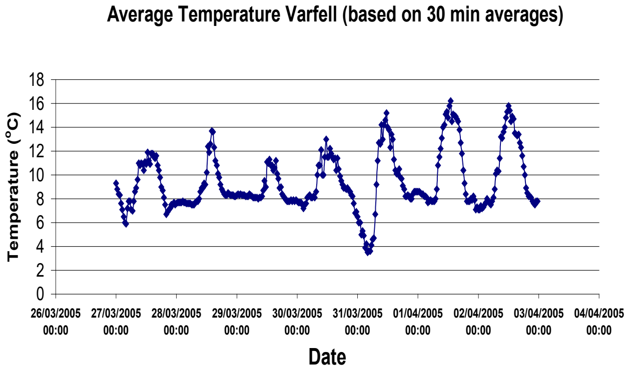


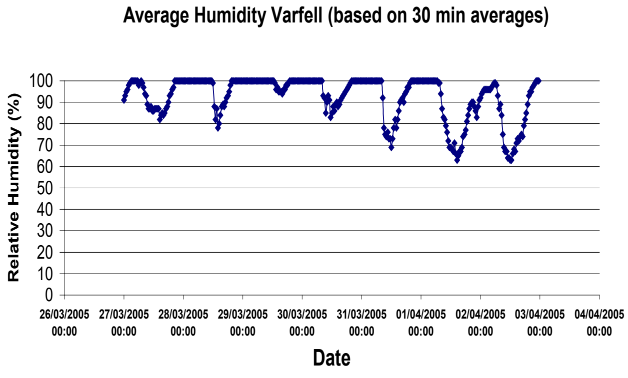


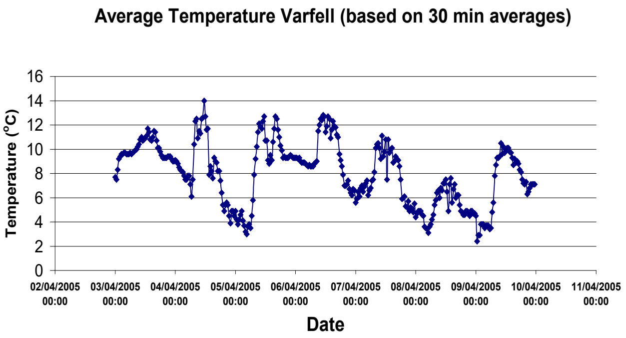








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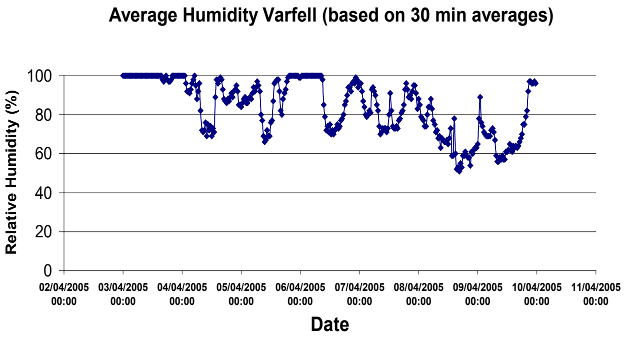
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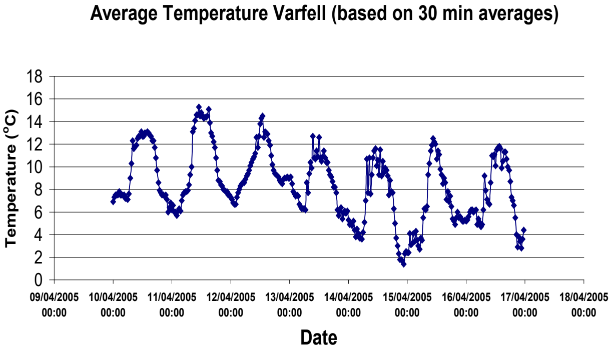
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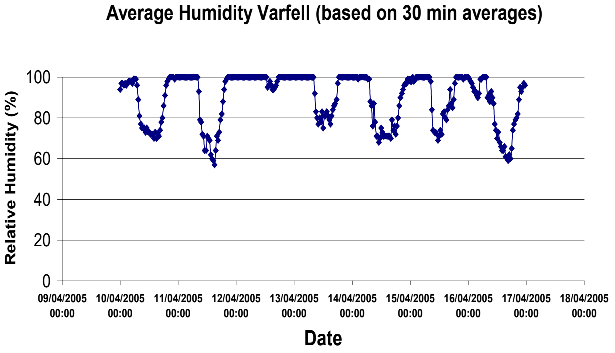
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Figure 4. Rainfall (▲) and leaf wetness (♦) at Nanceddan. Data averaged over 24-hour periods.

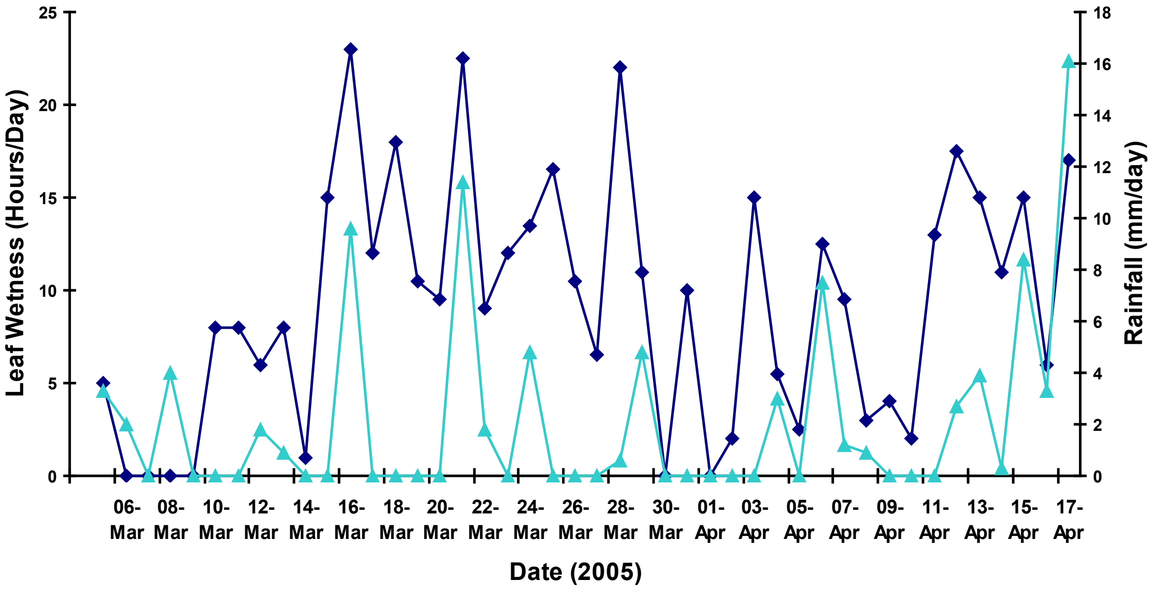

Figure 5. Predicted white mould infection score at Rosewarne. The infection score was averaged for 24-hour periods from 00:00 to 23:30 hours.

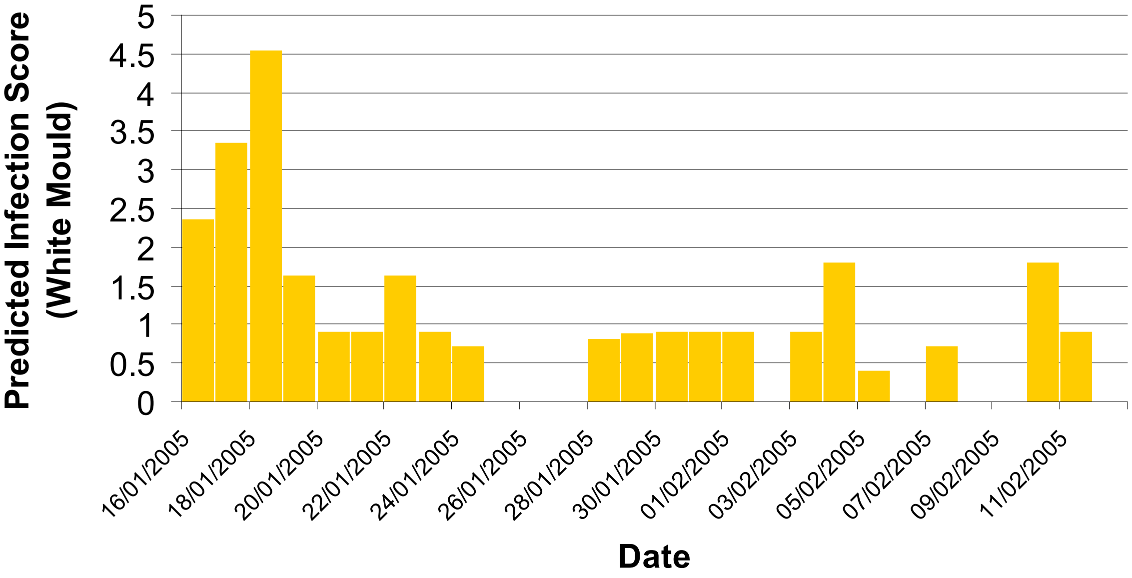

Figure 6. Predicted white mould infection score at Nanceddan. The infection score was averaged for 24-hour periods from 00:00 to 23:30 hours.

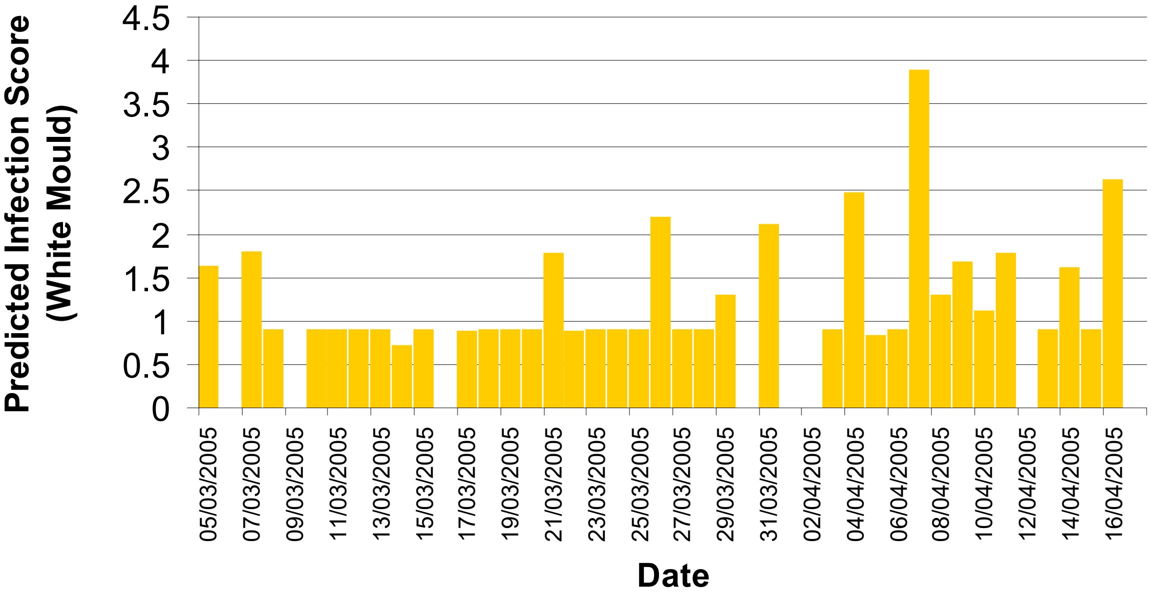

Figure 7. White mould monitoring at Rosewarne in 2005: number of plots showing white mould symptoms and incidence and severity scores.

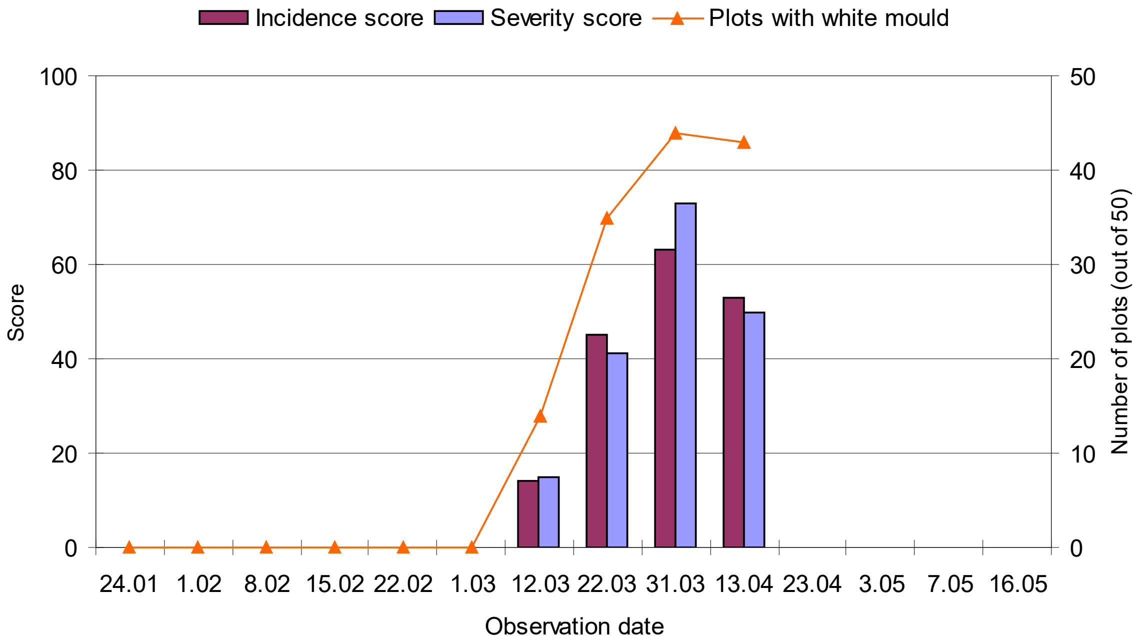

Figure 8. White mould monitoring at Nanceddan in 2005: number of plots showing white mould symptoms and incidence and severity scores. Note difference in left-hand scale between this Figure and Figure 5.