



Horticultural Fellowship Awards

Interim Report Form

Project title: Succession planning to sustain the UK's expertise
in field and laboratory plant pathology research
and development

Project number: CP 090

Project leader: Dr Angela Berrie
East Malling Research

Report: Annual report, September, 2012

Previous report: N/A

Fellowship staff: Dr Robert Saville
("Trainees")

Location of project: East Malling Research

Industry Representative: Andrew Tinsley,
Horticultural Development Company

Date project commenced: 7th November 2011

Date project completed
(or expected completion date): 6th November 2016

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AUTHENTICATION

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

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PROGRESS AGAINST OBJECTIVES

Objectives

Three fellowship projects (FP) were originally proposed in the work plan:

FP1 Sustainable control of storage rots of apple;

FP2 Exploring the extent of multi drug resistance (MDR) in *Botrytis cinerea* isolates in the UK population;

FP3 Exploring the possibility of a new pathogen, *Gnomonia fragariae*, affecting UK strawberry plantations.

The MDR project was considered to be outside the HDC remit and so the idea has been developed into a CRD proposal. The MDR fellowship project has been replaced with:

FP4 Evaluation of an apple mildew management programme through monitoring compared to a routine programme.

All fellowship projects are ongoing. In addition to specific fellowship projects, training has been provided through assistance and management of existing research projects and generic on-job training.

Formal training courses will be undertaken in years 2-3 including pesticide application training, identification of fungi of agricultural importance (IMI course) and BASIS training.

Objective	Original planned completion date	Actual completion date	Revised planned completion date
FP1 survey of 2011/12 storage season	01/05/2012	01/05/2012	
FP3 specific diagnostic and culturing techniques	01/10/2012	ongoing	
FP4 setup and monitoring trial (year 1)	01/10/2012	01/10/2012	

Assist in existing projects	ongoing		
Pesticide application course	07/11/2012		01/03/2013
Fungi identification course	07/11/2012		01/06/2012

Summary of Progress

All fellowship projects have commenced but by their nature they are on-going. The fellowship projects were designed to provide training across multiple crops (top fruit and soft fruit) and areas (fungal culturing and identification, chemical and biological control, growing systems and marketing) required for a plant pathologist.

Training has been supplemented through involvement in existing projects (listed in Appendix 1) and contribution to the development of future research projects and procurement of funds to support them.

Collaboration and networking with researchers, growers and industry representatives have complemented the training progress.

Milestones not being reached

Formal training courses have been postponed until years 2-3 because they have either not been available in 2012 (IMI fungal identification) or would be more beneficial having had some previous experience (Pesticide application and BASIS).

Do remaining milestones look realistic?

All other milestones have a realistic completion date

Other achievements in the last year not originally in the objectives

Development of proposals for the procurement of R & D funding (HDC tree fruit, CRD, TSB, BBSRC)

Working with colleagues within and outside of EMR to explore and develop future research areas (e.g. collaborations with Richard Harrison, Genetic Crop Improvement Programme, EMR, on translating advancements in genetic based resistance discovered in model plant species and agricultural crops and application of them to the breeding of horticultural/perennial crops and species.

Publication of peer reviewed articles in a scientific Journal (Saville et al., 2012, Journal of Experimental Botany, 63 (3): 1271-1283)

CHANGES TO PROJECT

Are the current objectives still appropriate for the Fellowship?

Other than the omission of the MDR Botrytis fellowship project and replacement with the mildew management trial as stated above no further changes need to be made.

GROWER SUMMARY

Headline

- A Horticultural Fellowship is dedicated to research into powdery mildew and storage rots of apple as well as a new pathogen of strawberry.

General background

Dr Angela Berrie has attained recognition for her role as the UK's leading field plant pathologist for fruit. With over 35 years of experience Dr Berrie's vast knowledge of plant pathology is respected by scientists and industry alike. Dr Berrie has greatly contributed to crop protection in the horticultural industry from applied field consultancy and plant clinic diagnostic skills to strategic research and development projects. A future gap in the application of these skills to UK horticulture has been identified as Dr Berrie approaches retirement. This horticultural fellowship has been created as part of the successional planning to sustain the UK's expertise in field and laboratory plant pathology research and development. Dr Robert Saville has been appointed as the Fellow to undertake this work and describes his work to date in this report.

Objectives

As part of the training fellowship three projects were proposed;

- (1) Sustainable control of storage rots of apple
- (2) Exploring the possibility of a new pathogen, *Gnomonia fragariae*, affecting UK strawberry plantations
- (3) Assessing the effectiveness of responsive apple mildew management through monitoring compared to a routine programme.

In addition to the fellowship projects above and assisting with existing projects at EMR, training has been provided in field and laboratory diagnostics.

This Grower Summary report is split into three sections summarising each objective of the fellowship.

Grower summary 1 - Sustainable control of storage rots of apple

Headline

- The incidence of *Gloeosporium* rots developing in store is increasing.

Background and expected deliverables

Types of storage rot

Fungal rots can result in significant losses in stored apples, particularly in fruit stored beyond January. Certain packhouses will record losses due to rots for individual bins of fruit, thus relating the loss to particular orchards, harvest time and pre-harvest factors. However they rarely identify the rots present. It is important to identify the rot profile in stored apples over time to build a dataset (including orchards, harvest time and pre-harvest factors) from which to base management strategies.

In previous surveys, *Nectria*, *Botrytis*, brown rot, *Penicillium*, *Phytophthora* and *Gloeosporium* have been identified as the main rots in apple. Other rots such as *Colletotrichum* sp, *Fusarium* sp, *Botryosphaeria* and *Phomopsis* have been increasing in incidence. A greater understanding of the epidemiology of these rots has helped in informing management strategies to reduce their prevalence. For example, *Nectria galligena*, the causal agent of apple canker, is also responsible for causing two distinct storage rot phenotypes; eye rot and cheek rot. Epidemiological studies have shown that fruit is most susceptible to *N. Galligena* at blossom and petal fall (Xu & Robinson, 2010) with successful infection leading to eye rot. Fruit susceptibility was shown to decline through summer and increase slightly near harvest, with infected fruit expressing *Nectria* cheek rot in store. In a Defra project HH3232STF it was shown that early season treatments targeted at *N. galligena* reduced the incidence of *Nectria* eye rot in store. Late season treatments will reduce the incidence of *Nectria* cheek rot, although issues of residues on fruit arising from treating close to harvest mean that early marketing of fruit, particularly in orchards with high levels of canker, is advisable.

Rot risk assessment

The concept of rot risk assessment was introduced via the Apple Best Practice Guide in 2001. The rot risk assessment takes account of various pre-harvest factors to predict the

level of rot likely to occur in store and thus inform a management strategy, be it pre-harvest treatments, selective picking or storage term, to minimise losses in store. The factors assessed pre-harvest are; daily rainfall, orchard factors, fungal inoculum (brown rot and canker), crop load, % bare ground (*Phytophthora*), % crop <0.5 metre from ground, orchard rot history and fruit storage potential (mineral composition and firmness). For example *Phytophthora* rot risk is influenced by three key factors; rainfall in 15 days prior to harvest, % bare ground and % crop <½ metre from ground.

In addition to rot risk assessment other management strategies can be employed to minimise losses in store such as selective picking. Only undamaged fruit is harvested and all fruit below 0.5 metres above the ground is excluded. This reduces the risk of introducing fungal rots, such as brown rot and *Penicillium* rot which establish on damaged fruit, and also *Phytophthora* rot which is prevalent on low hanging fruit, into the bin.

Pre-harvest fungicides applied for rot control are generally applied two to four weeks before harvest resulting in a high risk of residues in the fruit. By applying the recommendations set out in the rot risk assessment, such treatments could be avoided, reducing the risk of pesticide residues on fruit whilst reducing the financial and environmental costs of pesticide application.

In previous rot surveys, a trend has emerged of an increasing prevalence of *Gloeosporium* sp. Current research has not identified any clear factors on which risk assessment could be based apart from fruit set (light crop = risk). Decisions on risk are therefore mainly based on rot history and rainfall. As the trend of increasing losses due to *Gloeosporium* is likely to continue, it is important to increase our understanding of this storage rot and develop preventative measures to reduce rot prevalence.

Expected deliverables

The work in this objective is expected to determine which fungal rots are currently causing the greatest losses of fruit in store and to determine how the rot profile in store has changed over time since previous rot surveys. The work will also develop and evaluate management strategies for controlling rots.

Summary of the project and main conclusions

Five pack houses were visited in Kent weekly from January – March 2012. A visit was also made to a packhouse in Hereford in March. Rots were assessed on the grader of whatever

variety was being graded at the time of the visit. Rots were identified visually and numbers recorded.

Unidentified rots were cultured on to PDA and identified from spores or characteristic culture growth. Any isolates of *Gloeosporium* were collected to molecularly identify the species responsible for infection. Likewise *Botrytis* and *Fusarium* species have been collected for a fungicide resistance project and a *Fusarium* mycotoxin project respectively.

The levels of rainfall were very low during the spring of 2011 compared to the 50 year average.

Apple – Cox, Egremont Russet, Rosy Red

In the rot survey conducted in January-March 2012, rot levels were relatively low with an overall average loss of 2.4%, 2.5% and 2.0% for cvs. Cox, Egremont Russet and Rosy Red respectively. As has been observed in previous rot surveys, brown rot (caused by *Monolinia fructigena*) is the rot causing the highest losses, accounting for nearly 50% of the losses. *Gloeosporium*, *Nectria* and *Botrytis* are the next three most important rots to cause significant losses. *Phytophthora*, *Penicillium*, *Botryosphiria*, *Mucor* and *Phomopsis* were also recorded at low levels.

Apple – Bramley's Seedling

Losses due to rots in stored Bramley were the highest of all the cultivars assessed, with an average of 3% losses estimated. Losses increased with storage time with one sample, assessed in March in which losses were estimated at 7%. The main rots attributed to these losses were brown rot, *Penicillium*, *Phomopsis*, *Nectria* and *Fusarium*.

Apple – Braeburn, Gala, Jazz and Rubens

Negligible losses were recorded in stored Braeburn, Gala, Jazz and Rubens with an overall average loss of 0.3%, 0.5%, 0.3% and 0.1% respectively. For Braeburn, the main rots were brown rot and *Penicillium*. Losses in Gala were accounted to brown rot and *Nectria* and the main rot responsible for losses in Jazz was *Botrytis*.

Pear – Conference and Comice

Losses due to rots in pears overall was 2%. The main rots responsible for losses were *Botrytis* followed by brown rot. *Penicillium* and *Gloeosporium* were also present.

Conclusions

- Relatively low losses were recorded during the 2011/12 storage season which was probably on account of the dry spring of 2011.
- The 2012 growing season was very wet leading to a greater risk of rots developing in store.
- In general similar trends have been observed in the rot profile, most notably an increase in the incidence of *Gloeosporium*.
- A collection of *Gloeosporium* isolates has been assembled during the 2012 rot survey to be molecularly identified to determine the species level which will inform management strategy.

Financial benefits

- At this stage of the Fellowship project, no financial benefits for growers have yet been identified.

Action points for growers

- At this stage of the Fellowship project, no action points for growers have yet been identified.

Grower summary 2 – Exploring the possibility of a new pathogen, *Gnomonia fragariae*, affecting UK strawberry plantations

Headline

- Field samples examined in the first year exhibiting *G. fragariae* symptoms together with anecdotal evidence suggest that the pathogen is present in the UK and capable of causing disease.

Background and expected deliverables

Strawberry plays host to a large number of pathogens which can weaken and kill infected plants resulting in a reduction in a plantation's fruit yield and quality. Among these pathogens are a group which cause disease in the root system, crown and at the base of petioles. These diseases can cause serious damage to the host due to the disruption of the vascular system supplying the rest of the plant with water and nutrients. Such diseases include crown rot and red core caused by *Phytophthora cactorum* and *P. fragariae* respectively, Verticillium wilt and disorders caused by *Colletotrichum* sps.

It is these diseases which have contributed to the large scale adoption of the production systems used in modern strawberry production, whereby plantations are replanted on a yearly or two-yearly cycle rather than being a perennial crop. The emergence of new pathogen threats is increasing with an increase in worldwide trade of fresh produce and nursery stock and with the changing climate. It is important to identify these threats early so that diagnostics and control measures can be improved and implemented more rapidly. It is on this basis that investigations have been implemented on the presence of a new pathogen, *Gnomonia fragariae*, in UK strawberry plantations.

Gnomonia fragariae is an ascomycete fungus belonging to Diaporthales. *G. Fragariae* has long been considered as a saprophyte, colonising dead tissues of strawberry plants (Klebahn, 1918). However it has recently been shown to be the cause of a severe root rot and petiole blight of strawberry in Latvia and Sweden (Morocco *et al.* 2006). The disease causes a discolouration of rhizome tissues and crown, collapse of plants from one side, red or yellow coloured older leaves and bluish green colour of younger leaves. All of these symptoms are indicative of *Phytophthora* infection and therefore may have been misidentified as such in the past. In fact anecdotal evidence suggests that UK derived samples that show all the hallmarks of crown rot infection have failed to be confirmed using

standard traditional procedures (EMR Plant clinic samples, Dr Angela Berrie, pers. comm.). The diagnosis, and therefore detection of the presence of *G. fragariae* in UK strawberries, is further hindered by the slow growth of fungus in culture meaning colonies are often overgrown by faster-growing fungi.

Within this Fellowship, an investigation of the extent of *G. fragariae* in the UK will be made and robust and reliable diagnostic procedures developed in order to develop and provide advice on effective control measures to minimise detrimental effect on UK strawberry production. EMR is well placed to carry out such investigations as the plant clinic service receives ~200 samples per year for diagnosis and testing. Among these we can assess the level of *G. fragariae* in the field. EMR also hosts scientists with a lot of experience in diagnostics and field isolation of pathogens. A field survey will also be conducted once suitable sites have been identified.

Summary of the project and main conclusions

Strawberry samples submitted to the EMR plant clinic during 2012 underwent the standard diagnostic tests. Samples which looked characteristic of crown rot (i.e. wilted, bluish-green young leaves, all or part of the plant is dead or stunted, discoloured crowns and root rot in later stages) were tested using traditional diagnostic tests (floating crown and petiole tissue on water and assessing the presence of sporangia) and using molecular biology techniques (Lateral flow devices, Forsite Diagnostics, UK). Positives were reported as such whilst negatives were further analysed (below) for *G. fragariae*. Field samples which exhibited the symptoms characteristic of *G. fragariae* as described by Morocko (2006) have been collected from Home farm, East Malling.

Isolation methods have been adapted from Morocko *et al.* (2006). Stem bases, crowns and roots showing disease symptoms are surface sterilised in 1.25% sodium hypochlorite for two to three minutes, then washed three times in sterile distilled water and dried. Tissue at the leading edge of infection (i.e. the margin of healthy and diseased tissue) is plated on water agar media.

Infected material was also prepared as above for incubation in damp chambers (enclosed lunch boxes lined with damp tissue paper) to encourage the formation of fruiting bodies. Permutations on the methodology above include; inclusion of a paraquat soak when

preparing plant material, supplementing agar with paraquat, using different agars and incubation under UV light.

Results

The majority of the samples submitted to the plant clinic exhibiting symptoms resembling crown rot were confirmed to have been infected with *P. cactorum*. A single sample which had several characteristic symptoms of crown rot but a distinctive staining of the crown uncharacteristic of that caused by the disease, was shown to be negative for *Phytophthora* when tested with traditional and molecular diagnostics. The sample is suspected to be infected with *G. fragariae*.

Attempts were made to isolate the causal agent of the infected sample submitted to the plant clinic to confirm the presence of *G. fragariae*. Unfortunately culturing the fungus from the plant material on to agar has been unsuccessful due to other, faster growing, fungi outgrowing the suspected presence of *G. fragariae*. Infected material has been placed in a damp chamber and is being incubated under UV to encourage the development of fruiting bodies. At the time of writing this report no fruiting bodies were present.

Field samples resembling the symptoms caused by *G. fragariae* have been collected from a plantation at Home Farm, East Malling in September and at the time of writing, were being incubated under UV to encourage the development of fruiting bodies.

Discussion

Field and plant clinic samples exhibiting symptoms characteristic of a petiole/crown/root based disease have been examined. The samples were negative for *Phytophthora* sp. and, based on the symptoms are suspected to be infected with *G. fragariae*. To date attempts to isolate the pathogen from diseased material have been unsuccessful. Sites have been identified which are likely to contain plants infected with *G. fragariae* and will be visited in the second year of the project. Isolation techniques will continue to be developed and optimised.

Conclusions

- Field samples examined in the first year exhibiting *G. fragariae* symptoms together with anecdotal evidence suggest that the pathogen is present in the UK and capable of causing disease.
- Isolation of the fungus from infected material has not yet been achieved but will continue in year 2.

Financial benefits

- At this stage of the Fellowship project, no financial benefits for growers have yet been identified.

Action points for growers

- At this stage of the Fellowship project, no action points for growers have yet been identified.

Grower summary 3 – Assessing the effectiveness of responsive apple mildew management through monitoring compared to a routine programme

Headline

- In the first year of a mildew trial, a managed fungicide programme successfully reduced levels of apple powdery mildew.

Background and expected deliverables

Apple powdery mildew (*Podosphaera leucotricha*) is a major pathogen affecting apple production. The disease can severely reduce yield and quality (through russetting and cracking) particularly on susceptible cultivars such as Cox and Jonagold. Some cultivars, such as Golden Delicious, have increased tolerance to mildew epidemics, meaning that the disease is less detrimental to yield and quality. Breeding of cultivars with increased resistance and tolerance is possible, and such cultivars are utilised in organic production systems. However market forces mean that susceptible cultivars are still grown in conventional systems, meaning that growers are heavily reliant on chemical control to manage mildew epidemics.

The lifecycle of any disease is important to consider when developing effective management strategies particularly on a perennial crop such as apple. *Podosphaera leucotricha* over-winters as mycelium in the fruit and vegetative buds formed in the previous season. Therefore the level of disease at the end of the previous season will influence the epidemic in the following season. These buds emerge in spring, either as mildewed blossoms at pink bud or mildewed shoot tips at petal fall, as primary mildew. The inoculum from the primary mildew spreads to extending shoots to create a secondary mildew epidemic which, under favourable conditions, can infect leaves and produce sporulating mildew colonies in about four to five days. If the secondary mildew epidemic is high, fruit and vegetative buds are colonised as they develop and seal, and the fungus remains quiescent in the dormant buds until the following spring.

The key factor for effective control of a mildew epidemic in apple is to maintain primary mildew at a low level. This can be achieved throughout the season. Physical removal of mildewed blossoms and shoots may be necessary at the beginning of the season where

mildew levels are high. Keeping on top of the secondary mildew epidemic will reduce the following season's primary mildew epidemic and it is important to maintain chemical control throughout the season right from green cluster until vegetative growth ceases. However it is also important to adopt a flexible and responsive approach to chemical control, which can be achieved through disease monitoring. Monitoring is an important strategy in controlling a seasonal epidemic and potentially enables a grower to rationalise fungicide input and also alerts the grower to ineffectiveness of a particular product (either due to insufficient spray cover or the development of fungicide resistance in a local mildew population).

Advice to growers is available on how to effectively manage mildew through monitoring (for example the Apple Best Practice Guide). However as mildew epidemics are so detrimental to yield and quality of fruit, growers may be apprehensive about adopting such practices.

This trial is designed as a demonstration of effectiveness of implementing a managed programme, informed by monitoring compared to a routine programme, treated every 10-14 days with a varied programme of mildewicides. The management tools available are; choice of fungicide (eradicant or protectant), fungicide dose, spray interval and spray volume. The decisions will be based on; mildew incidence, growth stage and current weather.

Summary of the project and main conclusions

Site

Orchard EE190, Home Farm, East Malling. The orchard was planted in 1998 and consists of alternate rows of Royal Gala and Self Fertile Queen Cox. Tree spacing is 3.5m between rows and 1.75m between trees in the row. Two plots were marked out; Managed (blue) and Routine (red). Each plot consists of six rows of each cultivar with 29 trees in each row making each plot 0.19 ha in area

Treatments

The routine plot was treated with a standard fungicide programme sticking to a 10-14 day spray interval as detailed in the Science Section (Table 9). The frequency, dose, volume and choice of fungicide in the managed programme were determined by monitoring and are detailed in Table 9 of the Science Section of the report.

Monitoring

In the managed plot only 20 shoots were assessed weekly in June and through to July for the presence of secondary mildew by examining the top five leaves, starting with the first

fully expanded leaf. If mildew was present on any of the leaves the shoot was recorded as mildewed. The result is expressed as % mildewed shoots. Table 10 in the Science Section describes the guidelines for decisions on fungicide use in the managed programme based on secondary mildew assessments (Apple Best Practice Guide, HDC).

Assessments

Vegetative primary mildew was assessed as total number of shoots per tree and number with mildew on ten trees per cultivar per plot. The results are expressed as % mildewed shoots.

Secondary mildew was assessed as numbers of mildewed leaves in top five leaves per shoot, taking first fully unfurled leaf as leaf 1. Five shoots on 20 trees were assessed per cultivar per plot. Secondary mildew was assessed in July, August and September.

Fruit quality will be assessed at harvest. Five hundred fruit will be picked per cultivar per plot and scored for fruit russet (0-4 scale, where 0=no russet, 1=russet on calyx and stalk end, 2=russet on cheek of fruit, 3 = rough russet and 4=russet with cracking), weight of 100 fruit, and number and weight of fruit ≥ 65 mm.

Results

In the first year of this long-term trial mildew levels were very high. This is in part due to the minimal input of fungicides being applied to the experimental orchard in the years prior to this trial resulting in a build-up of over wintering inoculum, evident from the high levels of vegetative primary mildew recorded in the spring. Levels of primary vegetative mildew in the experimental orchard used in this trial averaged 15.3%. According to the Apple Best Practice Guide, levels of primary mildew on shoots greater than 2% indicate that controlling the mildew epidemic during the season will be problematic. In addition to the high level of inoculum at the beginning of the season the weather conditions through the growing season of 2012 were very favourable for mildew development and spread. Wet and warm weather provided high humidity for sporulation and allowed the trees to produce a lot of extension growth for colonisation.

Weekly monitoring during June reflected the high levels of mildew in the orchards which meant that the managed plot was sprayed with an intensive programme. An intensive programme was maintained due to the level of inoculum and the conditions being favourable for mildew through the growing season. By the end of July, monitoring revealed

that levels had dropped to 75% and 35% mildewed shoots for cvs. Gala and Cox respectively. Although still above the high disease rating threshold of 30%, the decision was made to revert to a routine programme for the managed plot due to the reduction in inoculum observed through monitoring and the reduction in extension growth.

A total of 11 mildewicide spray applications were made in the management plot with six-eight day intervals compared to seven sprays applied to the routine managed plot with 10-14 day spray intervals. The reduction in the mildew epidemic was evident in the management plot compared to the routine plot in both Cox and Gala.

High levels of mildew inoculum carried over from the previous season and highly favourable conditions for mildew spread made it very challenging to control the mildew epidemic in the trial plots in this single season. As a result of the high level of infection, the managed plot received an intensive programme of mildewicides. The managed plot, unsurprisingly, had a reduced mildew epidemic compared to the routine plot on all three assessment dates. This is the first year of a multi-year trial, and reductions in mildew levels are expected to accrete over successive seasons. Although fungicide inputs are initially higher, it is expected that once the mildew epidemic is under control fewer sprays will be required to achieve equivalent mildew control compared to the routine programme.

In order to fully benefit next year in 2013 from the reduction in the epidemic achieved in the managed plot this year, the orchard will continue to be monitored for shoot extension post harvest, which mild October weather can sometimes permit. This late extension growth can be colonised by residual mildew, often unchecked by fungicides, which then over-winters in terminal vegetative buds and emerges in the spring as primary mildew. Any late extension growth will trigger a further mildewicide application to the managed plot.

In addition to the mildew management trial commenced this year, an additional trial will be initiated to trial an integrated mildew management programme to include cultural control methods, such as the removal of mildewed tips, along with the inclusion of promising synthetic, alternative and biocontrol mildewicides identified in the apple mildew trial in the CP 77 / HL01109 SCEPTRE project.

Conclusions

- Having started the season with a very high level of inoculum. A reasonable level of control has been achieved through applying an intensive fungicide programme on the managed plot.
- The gains in a reduction in the mildew epidemic in the managed plot are expected to accrete in year 2 and subsequent years.

Financial benefits

- At this stage of the Fellowship project, no financial benefits for growers have yet been identified.

Action points for growers

- At this stage of the Fellowship project, no action points for growers have yet been identified.

SCIENCE SECTION

Introduction

General Background

Dr Berrie has attained recognition for her role as the UK's leading field plant pathologist for fruit. With over 35 years of experience Dr Berrie's vast knowledge of plant pathology is respected by scientists and industry alike. Dr Berrie has greatly contributed to crop protection in the horticultural industry from applied field consultancy and plant clinic diagnostic skills to strategic research and development projects. A future gap in the application of these skills to UK horticulture has been identified as Dr Berrie approaches retirement. This fellowship has been created as part of the successional planning to sustain the UK's expertise in field and laboratory plant pathology research and development.

Objectives

As part of the training fellowship three projects were proposed;

- (1) Sustainable control of storage rots of apple
- (2) Exploring the possibility of a new pathogen, *Gnomonia fragariae*, affecting UK strawberry plantations
- (3) Assessing the effectiveness of responsive apple mildew management through monitoring compared to a routine programme.

In addition to the fellowship projects above and assisting with existing projects at EMR, training was provided in field and laboratory diagnostics.

Sustainable control of storage rots of apple

Background

Types of storage rot

Fungal rots can result in significant losses in stored apples, particularly in fruit stored beyond January. Certain pack houses will record losses due to rots for individual bins of fruit, thus relating the loss to particular orchards, harvest time and pre-harvest factors, however they rarely identify the rots present. It is important to identify the rot profile in stored apples over time to build a dataset (including orchards, harvest time and pre harvest factors) from which to base management strategies.

In previous surveys *Nectria*, *Botrytis*, brown rot, *Penicillium*, *Phytophthora* and *Gloeosporium* have been identified as the main rots in apple. Other rots such as *Colletotrichum* sp, *Fusarium* sp, *Botryosphaeria* and *Phomopsis* have been increasing in

incidence. A greater understanding of the epidemiology of these rots has helped in informing management strategies to reduce their prevalence. For example, *Nectria galligena*, the causative agent of apple canker, is also responsible for causing two distinct storage rot phenotypes; eye rot and cheek rot. Epidemiological studies have shown that fruit is most susceptible to *N. Galligena* at blossom and petal fall (Xu & Robinson, 2010) with successful infection leading to eye rot. Fruit susceptibility was shown to decline through summer and increase slightly near harvest with infected fruit expressing Nectria cheek rot in store. In a Defra project HH3232STF it was shown that early season treatments targeted at *N. galligena* reduced the incidence of Nectria eye rot in store. Late season treatments will reduce the incidence of Nectria cheek rot, however issues of residues on fruit arising from treating close to harvest mean that early marketing of fruit, particularly in orchards with high levels of canker, is advisable.

Rot risk assessment

The concept of rot risk assessment was introduced via the Apple Best Practice Guide in 2001. The rot risk assessment takes account of various pre-harvest factors to predict the level of rot likely to occur in store and thus inform a management strategy, be it pre-harvest treatments, selective picking or storage term, to minimise losses in store. The factors assessed pre-harvest are; daily rainfall, orchard factors, fungal inoculum (brown rot and canker), crop load, % bare ground (*Phytophthora*), % crop <0.5 metre from ground, orchard rot history and fruit storage potential (mineral composition and firmness). For example Phytophthora rot risk is influenced by three key factors; rainfall in 15 days prior to harvest, % bare ground and % crop <½ metre from ground (Table 1).

Table 1. Factors influencing the risk of Phytophthora rot.

Factor	Criteria for risk
(1) Rainfall in 15 days prior to harvest	low or no rain = low risk 20 mm or >= high risk
(2) % bare ground	100% bare ground (overall herbicide) = high risk Overall grass or mulch or weed cover (0% bare ground) = low risk
(3) % crop <½ metre from ground	15% or >= risk

In addition to rot risk assessment other management strategies can be employed to minimise losses in store such as selective picking. Only undamaged fruit is harvested and all fruit below 0.5 metres above the ground is excluded. This reduces the risk of introducing fungal rots, such as brown rot and *Penicillium* rot which establish on damaged fruit, and also *Phytophthora* rot which is prevalent on low hanging fruit, into the bin.

Pre-harvest fungicides applied for rot control are generally applied two-four weeks before harvest resulting in a high risk of residues in the fruit. By applying the recommendations set out in the rot risk assessment such treatments could be avoided reducing the risk of pesticide residues on fruit whilst reducing the financial and environmental costs of pesticide application.

In previous rot surveys a trend has emerged of an increasing prevalence of *Gloeosporium* sp. Current research has not identified any clear factors on which risk assessment could be based apart from fruit set (light crop = risk). Decisions on risk are therefore mainly based on rot history and rainfall. As the trend of increasing losses due to *Gloeosporium* is likely to continue, it is important to increase our understanding of this storage rot and develop preventative measures to reduce rot prevalence.

Materials and methods

Five pack houses were visited in Kent weekly from January – March 2012. A visit was also made to a packhouse in Hereford in March (Table 2). Rots were assessed on the grader of whatever variety was being graded at the time of the visit. Rots were identified visually and numbers recorded.

Unidentified rots were cultured on to PDA and identified from spores or characteristic culture growth. Any isolates of *Gloeosporium* were collected to molecularly identify the species responsible for infection. Likewise *Botrytis* and *Fusarium* species have been collected for a fungicide resistance project and a *Fusarium* mycotoxin project respectively.

Table 2 Fruit Packhouses visited weekly in January – March 2012

Packhouse	Location	Number times visited
Newmafruit Farms Ltd	Howfield Farm, Chartham Hatch, Kent	5
F W Mansfield & Sons Ltd	Nickle Farm, Chartham, Kent	5
G Robertson	Breach Farm Goudhurst, Kent	5
Bardsley & Sons	River Farm, Staplehurst, Kent	4
J L Baxter & Son Ltd	Amsbury Farm, Hunton, Kent	5
Wye Fruit Ltd	Ledbury, Herefordshire	1

Results

Weather data for East Malling (Table 3) shows that the levels of rainfall were very low during the spring of 2011 compared to the 50 year average.

Apple – Cox, Egremont Russet, Rosy Red

In the rot survey conducted in January-March 2012 (Tables 4 and 7) rot levels were relatively low with an overall average loss of 2.4%, 2.5% and 2.0% for cvs. Cox, Egremont Russet and Rosy Red respectively. As has been observed in previous rot surveys brown rot (caused by *Monolinia fructigena*) is the rot causing the highest losses, accounting for nearly 50% of the losses. Gloeosporium, Nectria and Botrytis are the next three most important rots to cause significant losses. Phytophthora, Penicillium, Botryosphiria, Mucor and Phomopsis were also recorded at low levels.

Apple – Bramley's Seedling

Losses due to rots in stored Bramley (Table 5) were the highest of all the cultivars assessed, with an average of 3% losses estimated. Losses increased with storage time with one sample, assessed in March in which losses were estimated at 7%. The main rots attributed to these losses were brown rot, Penicillium, Phomopsis, Nectria and Fusarium.

Apple – Braeburn, Gala, Jazz and Rubens

Negligible losses were recorded in stored Braeburn, Gala, Jazz and Rubens (Tables 8 & 9) with an overall average loss of 0.3% 0.5%, 0.3% and 0.1% respectively. For Braeburn the main rots were brown rot and Penicillium. Losses in Gala were accounted to brown rot and Nectria and the main rot responsible for losses in Jazz was Botrytis.

Pear – Conference and Comice

Losses due to rots in pears overall was 2% (Table 8). The main rots responsible for losses were Botrytis followed by brown rot. Penicillium and Gloeosporium were also present.

Table 3. Monthly rainfall (mm) recorded at East Malling from March to September 2011. The 50 year average rainfall is presented for comparison.

Month	2011	50 year average
March	14.6	44.3
April	2.4	44.5
May	12.4	45.8
June	90.8	49.7
July	39.4	46.4
August	50.8	52
September	28.2	63.7

Table 4. Losses due to rots and rot incidence in apples cv. Cox assessed during grading in January-March 2012.

Cultivar	Packhouse	Date assessed	Date picked	Brown rot	Botrytis	Phytophthora	Penicillium	Nectria	Gloeosporium	Colletotrichum	Botryosphaeria	Mucor	Fusarium	Phomopsis	Alternaria	% loss estimated
Cox	The Breach	11/01/2012	08/09/2011	67	6	0	4	2	0	0	1	0	0	0	0	3-4%
	The Breach	18/01/2012	04/09/2011	85	0	2	0	7	11	0	0	0	0	0	0	<1%
	Amsbury	18/01/2012	08/09/2011	76	5	2	6	6	10	0	1	0	0	0	0	4.0%
	Mansfields	26/01/2012	31/08/2011	40	10	6	6	29	3	0	1	2	0	0	0	3.0%
	Mansfields	26/01/2012	?	7	14	5	2	0	25	0	0	0	0	0	0	1.0%
	Jan Mean			55.0	7.0	3.0	3.6	8.8	9.8	0.0	0.6	0.4	0.0	0.0	0.0	2.50%
	The Breach	01/02/2012	28/08/2011	39	9	0	3	9	15	0	0	0	0	0	0	3-4%
	Bardsley	01/02/2012	05/09/2011	30	0	0	1	7	0	0	0	0	0	0	0	0.5%
	Mansfields	02/02/2012	07/09/2011	15	3	0	2	0	2	0	0	0	0	0	0	<0.5%
	Bardsley	08/02/2012	05/09/2011	33	3	0	1	3	2	0	0	0	0	0	0	0.5%
	Mansfields	09/02/2012	05/09/2011	9	15	7	8	8	10	0	0	11	0	0	0	1.0%
	Mansfields	17/02/2012	05/09/2011	30	11	0	5	7	12	0	0	0	0	0	0	4-5%
	The Breach	22/02/2012	03/09/2011	53	6	0	5	38	16	0	0	0	0	0	0	8.0%
	Mansfields	24/02/2012	?	40	7	1	0	32	16	0	0	1	0	4	0	3.0%
	Mansfields	24/02/2012	03/09/2011	19	2	1	1	1	2	0	0	2	0	0	0	1.0%
	Feb Mean			29.8	6.2	1.0	2.9	11.7	8.3	0.0	0.0	1.6	0.0	0.4	0.0	2.60%
	Bardsley	01/03/2012	27/08/2011	12	1	0	4	0	0	0	2	0	0	0	0	<1%
	Mansfields	02/03/2012	?	8	6	3	3	20	16	0	0	0	0	0	0	2.0%
	Bardsley	07/03/2012	27/08/2011	29	0	0	4	5	6	0	0	0	0	0	0	<1%

	Wye Fruit	15/03/2012	12/09/2011	0	6	0	0	32	37	0	0	0	0	0	0	?
	Wye Fruit	15/03/2012	14/09/2011	0	0	0	0	7	55	0	0	0	0	0	0	?
	Wye Fruit	15/03/2012	13/09/2011	0	0	0	0	1	72	0	0	0	0	0	0	?
	Wye Fruit	15/03/2012	12/09/2011	0	1	0	1	64	16	0	0	0	1	2	0	?
	Bardsley	20/03/2012	?	56	30	0	1	22	1	0	3	0	0	0	0	2.0%
	NFF	22/03/2012	31/08/2011	47	17	0	5	8	56	0	0	0	0	0	0	4.00%
	March Mean			16.9	6.8	0.3	2.0	17.7	28.8	0.0	0.6	0.0	0.1	0.2	0.0	2.00%
	Overall Mean			30.2	6.6	1.2	2.7	13.4	16.7	0.0	0.3	0.7	0.0	0.3	0.0	

Table 5. Losses due to rots and rot incidence in apples cv. Bramley's Seedling assessed during grading in January-March 2012.

Cultivar	Packhouse	Date assessed	Date picked	Brown rot	Botrytis	Phytophthora	Penicillium	Nectria	Gloeosporium	Colletotrichum	Botryosphaeria	Mucor	Fusarium	Phomopsis	Alternaria	% loss estimated
Bramley	Mansfields	12/01/2012	22/08/2011	76	2	0	3	3	1	1	0	0	0	0	0	1-2%
	Mansfields	19/01/2012	19/08/2011	51	2	1	0	4	0	0	0	0	0		0	<0.5%
	Jan Mean			63.5	2.0	0.5	1.5	3.5	0.5	0.5	0.0	0.0	0.0	0.0	0.0	1.00%
	The Breach	01/02/2012	25/08/2011	4	2	0	6	4	0	0	0	0	9	3	0	1-2%
	The Breach	08/02/2012	10/08/2011	39	1	0	8	1	0	0	0	0	0	1	0	5.0%
	Mansfields	09/02/2012	24/08/2011	36	1	0	11	10	12	0	0	1	6	4	0	1.0%
	Mansfields	17/02/2012	02/09/2011	1	0	0	2	0	2	0	0	0	1	0	0	3.0%
	Feb Mean			20.0	1.0	0.0	6.8	3.8	3.5	0.0	0.0	0.3	4.0	2.0	0.0	2.60%
	The Breach	01/03/2012	17/08/2011	45		0	10	2	2	0	1	1	5	0	0	3.0%
	Mansfields	02/03/2012	17/08/2011	4	0	0	1	1	1	0	0	0	2	3	0	3.0%
	The Breach	07/03/2012	17/08/2011	53	4	0	4	5	1	0	1	4	5	3	0	7.0%

	Mansfields	08/03/2012	09/09/2011	32	0	0	3	0	2	0	2	0	2	28	0	4.0%
	The Breach	20/03/2012	24/08/2011	53	1	0	3	10	2	0	0	0	8	4	0	<1%
	March Mean			37.4	1.3	0.0	4.2	3.6	1.6	0.0	0.8	1.0	4.4	7.6	0.0	3.60%
	Overall Mean			35.8	1.3	0.1	4.6	3.6	2.1	0.1	0.4	0.5	3.5	4.6	0.0	

Table 6. Losses due to rots and rot incidence in apples cv. Braeburn assessed during grading in January-March 2012.

Cultivar	Packhouse	Date assessed	Date picked	Brown rot	Botrytis	Phytophthora	Penicillium	Nectria	Gloeosporium	Colletotrichum	Botryosphaeria	Mucor	Fusarium	Phomopsis	Alternaria	% loss estimated
Braeburn	The Breach	11/01/2012	10/10/2011	2	9	2	1	0	1	0	0	0	0	0	0	<0.5%
	Mansfields	12/01/2012	20/10/2011	19	12	1	28	0	0	0	0	0	0	0	0	<0.1%
	Bardsley	18/01/2012	??/10/2011	35	0	0	2	0	0	0	0	0	0	0	0	<0.1%
	Mansfields	19/01/2012	18/10/2011	18	4	3	23	0	2	0	0	0	0	0	0	<0.1%
	Mansfields	19/01/2012	08/10/2011	28	9	0	12	0	3	0	0	0	1	0	0	<0.1%
	Mansfields	26/01/2012	17/10/2011	1	21	1	27	0	3	0	0	0	0	0	0	<0.1%
	Jan Mean			17.2	9.2	1.2	15.5	0.0	1.5	0.0	0.0	0.0	0.2	0.0	0.0	0.20%
	Mansfields	17/02/2012	01/11/2011	15	7	2	8	3	15	0	0	31	0	0	0	1.0%
	Bardsley	22/02/2012	30/09/2011	32	14	0	0	0	0	0	0	1	0	0	0	<0.1%
	Mansfields	24/02/2012	21/10/2011	2	6	0	32	21	3	0	0	0	0	0	0	0.5%
	NFF	24/02/2012	12/10/2011	22	9	1	11	4	14	0	0	0	0	0	0	0.10%
	Feb Mean			17.8	9.0	0.8	12.8	7.0	8.0	0.0	0.0	8.0	0.0	0.0	0.0	0.40%
	Mansfields	02/03/2012	10/10/2011	5	12	8	14	3	7	0	0	0	0	0	0	<0.1%
	Amsbury	07/03/2012	11/10/2011	7	5	0	6	0	4	0	0	0	0	0	0	0.1%
	Mansfields	08/03/2012	14/10/2011	1	0	0	13	3	22	0	0	0	0	0	0	0.1%
	NFF	08/03/2012	05/10/2011	21	9	2	4	5	7	0	0	0	0	0	0	0.50%
	Wye Fruit	15/03/2012	?	1	1	0	4	0	1	0	0	0	0	0	0	<0.1%
	Mansfields	22/03/2012	24/10/2011	7	6	1	10	2	6	0	0	0	0	1	0	0.1%
	March			7.0	5.5	1.8	8.5	2.2	7.8	0.0	0.0	0.0	0.0	0.2	0.0	0.20%

	Mean															
	Overall Mean			13.5	7.8	1.3	12.2	2.6	5.5	0.0	0.0	2.0	0.1	0.1	0.0	

Table 7. Losses due to rots and rot incidence in apples cvs. Gala, Jazz, Egremont Russet, Rosy Red and Rubens assessed during grading in January-March 2012.

Cultivar	Packhouse	Date assessed	Date picked	Brown rot	Botrytis	Phytophthora	Penicillium	Nectria	Gloeosporium	Colletotrichum	Botryosphaeria	Mucor	Fusarium	Phomopsis	Alternaria	% loss estimated
Gala	Mansfields	19/01/2012	08/09/2011	22	3	1	3	21	4	0	0	0	0	0	0	?
	Mansfields	26/01/2012	22/09/2011	55	7	1	11	0	2	0	0	0	0	0	0	0.5%
	Mansfields	02/02/2012	25/09/2011	24	15	7	7	5	7	0	0	0	0	0	0	0.5%
	Mansfields	09/02/2012	25/09/2011	39	12	0	4	16	12	0	0	0	0	0	0	0.5%
	NFF	09/02/2012	15/09/2011	18	1	0	8	11	11	0	0	0	0	0	0	0.10%
	Amsbury	22/02/2012	19/09/2011	10	2	0	11	28	12	0	0	2	0	0	0	1.0%
	Mansfields	02/03/2012	14/09/2011	18	2	0	5	15	15	0	0	0	0	0	0	0.1%
	Wye Fruit	15/03/2012	20/09/2011	0	1	0	0	5	2	0	0	0	0	0	0	?
	Wye Fruit	15/03/2012	19/09/2011	0	1	0	0	50	11	0	0	0	0	0	0	?
	Overall Mean			20.7	4.9	1.0	5.4	16.8	8.4	0.0	0.0	0.2	0.0	0.0	0.0	0.50%
Jazz	Amsbury	11/01/2012	10/10/2011	1	2	0	5	1	4	0	0	0	0	0	0	<0.1%
	Mansfields	12/01/2012	10/11/2011	1	0	25	2	0	0	0	0	0	0	0	0	<0.1%
	Mansfields	02/02/2012	13/10/2011	2	20	4	9	1	1	0	0	0	0	0	0	0.1%
	NFF	02/02/2012	10/10/2011	1	11	0	7	4	7	0	0	0	0	0	0	0.10%
	Mansfields	17/02/2012	13/10/2011	2	7	1	5	25	12	0	0	0	0	0	0	1-2%

	NFF	17/02/2012	27/09/2011	0	6	0	3	3	14	0	0	0	0	0	0	0.10%
	Mansfields	08/03/2012	11/10/2011	4	36	0	14	0	0	0	1	0	0	0	0	0.1%
	Overall Mean			1.6	11.7	4.3	6.4	4.9	5.4	0.0	0.1	0.0	0.0	0.0	0.0	0.30%
Egremont Russet	NFF	02/02/2012	08/09/2011	84	3	0	9	4	8	0	0	0	2	0	0	1%
	Amsbury	20/03/2012	16/09/2011	18	1	0	2	9	44	0	0	0	0	0	0	4.0%
	Overall Mean			51.0	2.0	0.0	5.5	6.5	26.0	0.0	0.0	0.0	1.0	0.0	0.0	2.50%
Rosy Red	Amsbury	01/03/2012	?	19	0	0	4	0	9	0	1	0	0	0	0	2.0%
Rubens	Bardsley	11/01/2012	25/09/2011	24	6	1	9	1	5	0	1	0	1	0	0	0.1%

Table 8. Losses due to rots and rot incidence in Pear cvs. Comice and Conference assessed during grading in January-March 2012.

Cultivar	Packhouse	Date assessed	Date picked	Brown rot	Botrytis	Phytophthora	Penicillium	Nectria	Gloeosporium	Colletotrichum	Botryosphaeria	Mucor	Fusarium	Phomopsis	Alternaria	% loss estimated
Comice	Amsbury	01/02/2012	13/09/2011	6	39	0	1	2	3	0	0	0	0	0	0	5.0%
	Amsbury	08/02/2012	12/09/2011	8	29	0	1	0	0	0	0	0	0	0	0	1.0%
	Overall Mean			7.0	34.0	0.0	1.0	1.0	1.5	0.0	0.0	0.0	0.0	0.0	0.0	3.00%
Conference	Mansfields	19/01/2012	22/08/2011	11	34	1	7	0	2	0	0	0	0	0	2	<0.1%
	Mansfields	26/01/2012	22/08/2011	4	10	0	5	0	0	0	0	0	0	0	0	1-2%
	Mansfields	02/02/2012	29/08/2011	20	11	0	2	0	1	0	0	0	0	0	0	0.5%
	NFF	09/02/2012	15/08/2011	17	18	0	3	1	0	0	0	0	0	0	0	0.50%
	NFF	17/02/2012	05/09/2011	20	44	0	10	0	1	0	1	4	0	0	0	<1%

	NFF	22/03/2012	27/08/2011	13	10	0	3	8	4	0	0	0	0	0	0	2.00%
	Overall Mean			14.2	21.2	0.2	5.0	1.5	1.3	0.0	0.2	0.7	0.0	0.0	0.3	0.90%

Discussion

In the 2012 rot survey conducted in this study, rot incidence was relatively low compared with previous surveys. Weather data for East Malling (Table 3) shows that the levels of rainfall were very low during the spring of 2011 compared to the 50 year average and, although June rainfall in 2011 was nearly double that of the 50 year average for June, this coincides with a period of reduced susceptibility to rot-causing pathogens. In general, the 2012 rot survey followed similar trends to the surveys conducted in recent years. Most losses were due to brown rot (*Monilinia fructigena*). Newer cultivars, such as Braeburn, Jazz and Rubens had exceptionally low losses (0.1-0.3%) compared to Cox and Bramley (2.4-3%); and the incidence of *Gloeosporium* in stored apples increased following the trend of the last four years.

Gloeosporium is one of the most important causes of losses in stored apple in other parts of Europe. Recent increases in incidence in the UK may reflect changes in climate. It is important to gain a better understanding of this rot in order to curb the upward trend of incidence. To this end a collection of *Gloeosporium* isolates has been made from the 2012 rot survey and these are to be molecularly identified to the species level. Through this exercise it is hoped that better control measures and recommendations can be developed.

The data from this current study, together with previous rot surveys conducted between 1995 and 2000 and 2008 to 2011, provide a useful data set of how the rot profile has changed over time and, when coupled with weather data and step changes in orchard treatments and storage protocols, provides a useful tool for developing and evaluating management strategies for controlling rots and identifying rots which are increasing in incidence (such as *Gloeosporium*).

East Malling Research has a long and successful history of developing optimal storage protocols for apple and pear cultivars and the Produce Quality Centre (PQC) continues this work today. Currently the PQC is working on several trials to incorporate the use of new technologies, such as SmartFresh™, into storage regimes. SmartFresh™ technology delays the ripening/senescence process controlled by ethylene whilst fruit is in store. The product's active ingredient, 1-methylcyclopropene (1-MCP) effectively mimics ethylene enabling it to interact with the ethylene receptors of the fruits and block their action. A reduction in the rate of ripening/senescence afforded by SmartFresh™ enables storage potential to be extended and/or other storage parameters such as temperature and CA, to be revised.

Ethylene is a key hormone involved in plant resistance responses therefore blocking ethylene signalling would be expected to have adverse effects on resistance to rot development. Preliminary data from PQC trials show that SmartFresh™ has a limited effect on rot development (Dr Debbie Rees, PQC/NRI pers. comm.), however further work is required. As the storage protocols developed at the PQC are adopted by commercial stores it will be important to analyse the effects on rot incidence. Therefore, future rot surveys will take into account storage protocols (i.e. + or - SmartFresh™, temperature and CA, if different from standard).

The 2012 growing season has been very wet throughout, leading to a greater risk of rot development in store. Prolonged wet periods during flowering, again in June and July and wet weather at the end of the season and into harvest mean that many of the rots are likely to increase in incidence, particularly *Phytophthora*, *Nectria* and *Botrytis* which are favoured by wet weather at different times in the growing season. The rot survey will continue next year.

Conclusions

- Relatively low losses were recorded during the 2011/12 storage season which was probably on account of the dry spring of 2011.
- The 2012 growing season has been very wet leading to a greater risk of rots developing in store.
- In general similar trends have been observed in the rot profile, most notably an increase in the incidence of *Gloeosporium*.
- A collection of *Gloeosporium* isolates has been assembled during the 2012 rot survey to be molecularly identified to determine the species level which will inform management strategy.

1. Exploring the possibility of a new pathogen, *Gnomonia fragariae*, affecting UK strawberry plantations

Background

Strawberry plays host to a large number of pathogens which can weaken and kill infected plants resulting in a reduction in a plantation's fruit yield and quality. Among these pathogens are a group which cause disease in the root system, crown and at the base of petioles. These diseases can cause serious damage to the host due to the disruption of the vascular system supplying the rest of the plant with water and nutrients. Such diseases include crown rot and red stele caused by *Phytophthora cactorum* and *P. fragariae* respectively, Verticillium wilt and disorders caused by *Colletotrichum* sps.

It is these diseases which have contributed to the large scale adoption of the production systems used in modern strawberry production, whereby plantations are replanted on a yearly or two-yearly cycle rather than being a perennial crop. The emergence of new pathogen threats is increasing with an increase in worldwide trade of produce and nursery stock and with the changing climate. It is important to identify these threats early so that diagnostics and control measures can be improved and implemented more rapidly. It is on this basis that investigations have been implemented on the presence of a new pathogen, *Gnomonia fragariae*, in UK strawberry plantations.

Gnomonia fragariae is an ascomycete fungus belonging to Diaporthales. *G. Fragariae* has long been considered as a saprophyte, colonising dead tissues of strawberry plants (Klebahn, 1918). However it has recently been shown to be the cause of a severe root rot and petiole blight of strawberry in Latvia and Sweden (Morocco *et al.* 2006). The disease causes a discolouration of rhizome tissues and crown, collapse of plants from one side, red or yellow coloured older leaves and bluish green colour of younger leaves. All of these symptoms are indicative of *Phytophthora* infection and therefore may have been misidentified as such in the past. In fact anecdotal evidence suggests that UK derived samples that show all the hallmarks of crown rot infection have failed to be confirmed using standard traditional procedures (EMR Plant clinic samples, Dr Angela Berrie, pers. comm.). The diagnosis, and therefore detection of the presence of *G. fragariae* in UK strawberries, is further hindered by the slow growth of fungus in culture meaning colonies are often overgrown by faster-growing fungi.

An investigation of the extent of *G. fragariae* is required in the UK and robust and reliable diagnostic procedures developed in order to develop and provide advice on effective control

measures to minimise detrimental effect on UK strawberry production. EMR are well placed to carry out such investigations as the plant clinic service receives ~200 samples per year for diagnosis and testing. Among these we can assess the level of *G. fragariae* in the field. EMR also hosts scientists with a lot of experience in diagnostics and field isolation of pathogens. A field survey will also be conducted once suitable sites have been identified.

Materials and methods

Strawberry samples submitted to the EMR plant clinic during 2012 have undergone the standard diagnostic tests. Samples which looked characteristic of crown rot (i.e. wilted, bluish-green young leaves, all or part of the plant is dead or stunted, discoloured crowns and root rot in later stages) were tested using traditional diagnostic tests (floating crown and petiole tissue on water and assessing the presence of sporangia) and using molecular biology techniques (Lateral flow devices, Forsite Diagnostics, UK). Positives were reported as such whilst negatives were further analysed (below) for *G. fragariae*. Field samples which exhibited the symptoms characteristic of *G. fragariae* (Fig. 1) as described by Morocco (2006) have been collected from Home farm, East Malling.

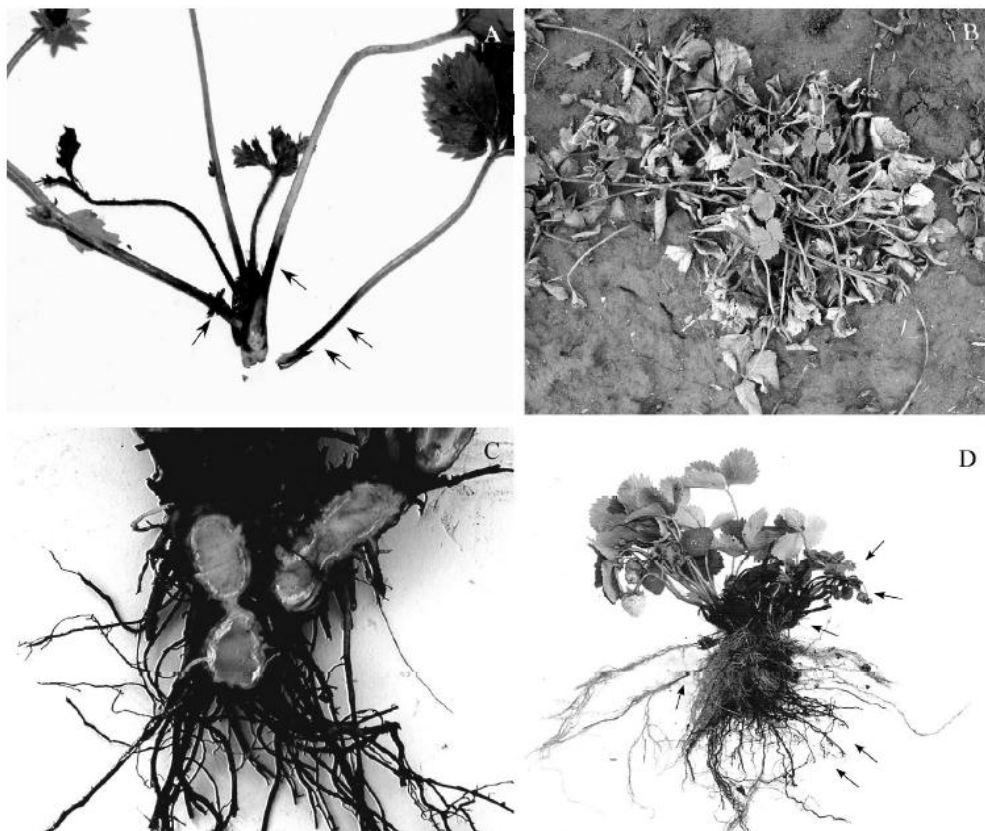


Fig. 1 Typical disease symptoms of strawberry plants infected with *G. fragariae* in the field: (A) black rot on petiole bases; (B) collapsed outer leaves; (C) black root rot and discoloration in crown; (D) severe stunt, black rot on roots and collapse of plant from one side. Figure from Morocco, 2006, Doctoral thesis.

Isolation methods have been adapted from Morocco *et al.* (2006). Stem bases, crowns and roots showing disease symptoms are surface sterilised in 1.25% sodium hypochlorite for 2-3 minutes then washed three times in sterile distilled water and dried. Tissue at the leading edge of infection (i.e. the margin of healthy and diseased tissue) is plated on water agar media.

Infected material was also prepared as above for incubation in damp chambers (enclosed lunch boxes lined with damp tissue paper) to encourage the formation of fruiting bodies. Permutations on the methodology above include; inclusion of a paraquat soak when preparing plant material, supplementing agar with paraquat, using different agars and incubation under UV light.

Results

The majority of the samples submitted to the plant clinic exhibiting symptoms resembling crown rot were confirmed to have been infected with *P. cactorum*. A single sample which had several characteristic symptoms of crown rot (Fig. 2a) but a distinctive staining of the crown (Fig. 2b) uncharacteristic of that caused by the disease, was shown to be negative for Phytophthora when tested with traditional and molecular diagnostics. The sample is suspected to be infected with *G. fragariae*.

Attempts were made to isolate the causative agent of the infected sample submitted to the plant clinic to confirm the presence of *G. fragariae*. Unfortunately culturing the fungus from the plant material on to agar has been unsuccessful due to other, faster growing, fungi outgrowing the suspected presence of *G. fragariae*. Infected material has been placed in a damp chamber and is being incubated under UV to encourage the development of fruiting bodies. At the time of writing this report no fruiting bodies were present.

Field samples resembling the symptoms caused by *G. fragariae* have been collected from a plantation at Home Farm, East Malling in September and are currently being incubated under UV to encourage the development of fruiting bodies.



Fig. 2. Field (a) and crown (b) symptoms of suspected *G. fragariae* infected strawberry plants submitted to EMR plant clinic for diagnosis.

Discussion

Field and plant clinic samples exhibiting symptoms characteristic of a petiole/crown/root based disease have been examined. The samples were negative for *Phytophthora* sp. and, based on the symptoms are suspected to be infected with *G. fragariae*. To date attempts to isolate the pathogen from diseased material have been unsuccessful. Sites have been identified which are likely to contain plants infected with *G. fragariae* and will be visited in the second year of the project. Isolation techniques will continue to be developed and optimised.

Conclusions

- Field samples examined in the first year exhibiting *G. fragariae* symptoms together with anecdotal evidence suggest that the pathogen is present in the UK and capable of causing disease.

- Isolation of the fungus from infected material has not yet been achieved but will continue in year 2.

2. Assessing the effectiveness of responsive apple mildew management through monitoring compared to a routine programme.

Background

Apple powdery mildew (*Podosphaera leucotricha*) is a major pathogen affecting apple production. The disease can severely reduce yield and quality (through russetting and cracking) particularly on susceptible cultivars such as Cox and Jonagold. Some cultivars, such as Golden Delicious, have increased tolerance to mildew epidemics, meaning that the disease is less detrimental to yield and quality. Breeding of cultivars with increased resistance and tolerance is possible, and such cultivars are utilised in organic production systems, however market forces mean that susceptible cultivars are still grown in conventional systems, meaning that growers are heavily reliant on chemical control to manage mildew epidemics.

The lifecycle of any disease is important to consider when developing effective management strategies particularly on a perennial crop such as apple. *Podosphaera leucotricha* over-winters as mycelium in the fruit and vegetative buds formed in the previous season, therefore the level of disease at the end of the previous season will influence the epidemic in the following season. These buds emerge in spring, either as mildewed blossoms at pink bud or mildewed shoot tips at petal fall, as primary mildew. The inoculum from the primary mildew spreads to extending shoots to create a secondary mildew epidemic which, under favourable conditions, can infect leaves and produce sporulating mildew colonies in about 4-5 days. If the secondary mildew epidemic is high, fruit and vegetative buds are colonised as they develop and seal, and the fungus remains quiescent in the dormant buds until the following spring.

The key factor for effective control of a mildew epidemic in apple is to maintain primary mildew at a low level. This can be achieved throughout the season. Physical removal of mildewed blossoms and shoots may be necessary at the beginning of the season where

mildew levels are high. Keeping on top of the secondary mildew epidemic will reduce the following season's primary mildew epidemic and it is important to maintain chemical control throughout the season right from green cluster until vegetative growth ceases. However it is also important to adopt a flexible and responsive approach to chemical control, which can be achieved through disease monitoring. Monitoring is an important strategy in controlling a seasonal epidemic and potentially enables a grower to rationalise fungicide input and also alerts the grower to ineffectiveness of a particular product (either due to insufficient spray cover or the development of fungicide resistance in a local mildew population).

Advice to growers is available on how to effectively manage mildew through monitoring (for example the Apple Best Practice Guide), however as mildew epidemics are so detrimental to yield and quality of fruit, growers may be apprehensive of adopting such practices. This trial is designed as a demonstration of effectiveness of implementing a managed programme, informed by monitoring compared to a routine programme, treated every 10-14 days with a varied programme of mildewicides. The management tools available are; choice of fungicide (eradicant or protectant), fungicide dose, spray interval and spray volume. The decisions will be based on; mildew incidence, growth stage and current weather.

Materials and methods

Site

Orchard EE190, Home Farm, East Malling. The orchard was planted in 1998 and consists of alternate rows of Royal Gala and Self Fertile Queen Cox. Tree spacing is 3.5m between rows and 1.75m between trees in the row. Two plots were marked out; Managed (blue) and Routine (red). Each plot consists of six rows of each cultivar with 29 trees in each row making each plot 0.19 ha in area

Treatments

The routine plot was treated with a standard fungicide programme sticking to a 10-14 day spray interval as detailed in Table 9. The frequency, dose, volume and choice of fungicide in the managed programme were determined by monitoring and are detailed in Table 9.

Table 9. Treatment programmes applied to routine and managed plots through trial period.

Treatment Number	Date Applied	Product	Dose Rate/ha	Concentration	Routine (RED)	Managed (Blue)
1	25/05/2012	UKA384c	0.5L	2.5ml	Y ¹	Y
2	06/06/2012	Topas	0.5L	2.5ml	Y	Y
3	14/06/2012	Topas	0.5L	2.5ml		Y
4	18/06/2012	Cosine	0.5L	2.5ml	Y	Y
5	25/06/2012	Sulphur	5L	10ml		Y
		Potassium Bicarb	5kg	10g		
6	05/07/2012	Topas	0.5L	2.5ml	Y	Y
7	10/07/2012	Cosine	0.5L	2.5ml		Y
8	18/07/2012	Systhane	0.33L	1.65ml	Y	Y
		Bellis	0.8kg	4g		
9	23/07/2012	Topas	0.5L	2.5ml		Y
10	02/08/2012	Systhane	0.33L	1.65ml	Y	Y
		Bellis	0.8kg	4g		
11	21/08/2012	Topas	0.5L	2.5ml	Y	Y

¹ Y denotes treatment was applied in plot

Monitoring

In the managed plot only 20 shoots were assessed weekly in June and through to July for the presence of secondary mildew by examining the top five leaves, starting with the first fully expanded leaf. If mildew was present on any of the leaves the shoot was recorded as mildewed. The result is expressed as % mildewed shoots. Table 10 describes the guidelines for decisions on fungicide use in the managed programme based on secondary mildew assessments (Apple Best Practice Guide, HDC).

Table 10. Guidelines for decisions on fungicide use based on secondary mildew monitoring assessments. Adapted from the Apple Best Practice Guide, HDC

Disease rating	Mildewed shoots (%)	Action after petal fall
Low	<10	In cool weather with rainy spells or shoot growth is slow, opportunity to reduce fungicide by reducing dose (minimum dose = 25%) or extending spray interval.
Moderate	10-30	Maintain control. Consider improving programme by reducing spray interval or increasing fungicide dose (not exceeding label maximum) especially if weather is warm and humid and shoot growth rapid.
Potentially high	>30	Improve control immediately especially if shoots are growing, irrespective of weather. Shorten spray interval, increase fungicide dose (not exceeding label maximum), possibly increase spray volume. Consider changing fungicide. Check sprayer is working correctly.

Assessments

Vegetative primary mildew was assessed as total number of shoots per tree and number with mildew on ten trees per cultivar per plot. The results are expressed as % mildewed shoots.

Secondary mildew was assessed as numbers of mildewed leaves in top five leaves per shoot, taking first fully unfurled leaf as leaf 1. Five shoots on 20 trees were assessed per cultivar per plot. Secondary mildew was assessed in July, August and September.

Fruit quality will be assessed at harvest. Five hundred fruit will be picked per cultivar per plot and scored for fruit russet (0-4 scale, where 0=no russet, 1=russet on calyx and stalk end, 2=russet on cheek of fruit, 3 = rough russet and 4=russet with cracking), weight of 100 fruit, and number and weight of fruit ≥ 65 mm.

Results

In the first year of this long-term trial mildew levels were very high. This is in part due to the minimal input of fungicides being applied to the experimental orchard in the years prior to this trial resulting in a build-up of over wintering inoculum, evident from the high levels of vegetative primary mildew recorded in the spring (Table 11). Levels of primary vegetative mildew in the experimental orchard used in this trial averaged 15.3%. According to the Apple Best Practice Guide, levels of primary mildew on shoots greater than 2% indicate that controlling the mildew epidemic during the season will be problematic. In addition to the high level of inoculum at the beginning of the season the weather conditions through the growing season of 2012 were very favourable for mildew development and spread. Wet and warm weather provided high humidity for sporulation and allowed the trees to produce a lot of extension growth for colonisation.

Table 11. Vegetative primary mildew expressed as % mildewed shoots

Plot	Cultivar	
	Gala	Cox
Managed	11.8%	23.0%
Routine	13.0%	13.4%

Weekly monitoring during June reflected the high levels of mildew in the orchards (95-100% mildewed shoots; Fig 3) which meant that the managed plot was sprayed with an intensive programme. An intensive programme was maintained due to the level of inoculum and the conditions being favourable for mildew through the growing season. By the end of July monitoring revealed that levels had dropped to 75% and 35% mildewed shoots for cvs. Gala and Cox respectively. Although still above the high disease rating threshold of 30%, the decision was made to revert to a routine programme for the managed plot due to the reduction in inoculum observed through monitoring and the reduction in extension growth.

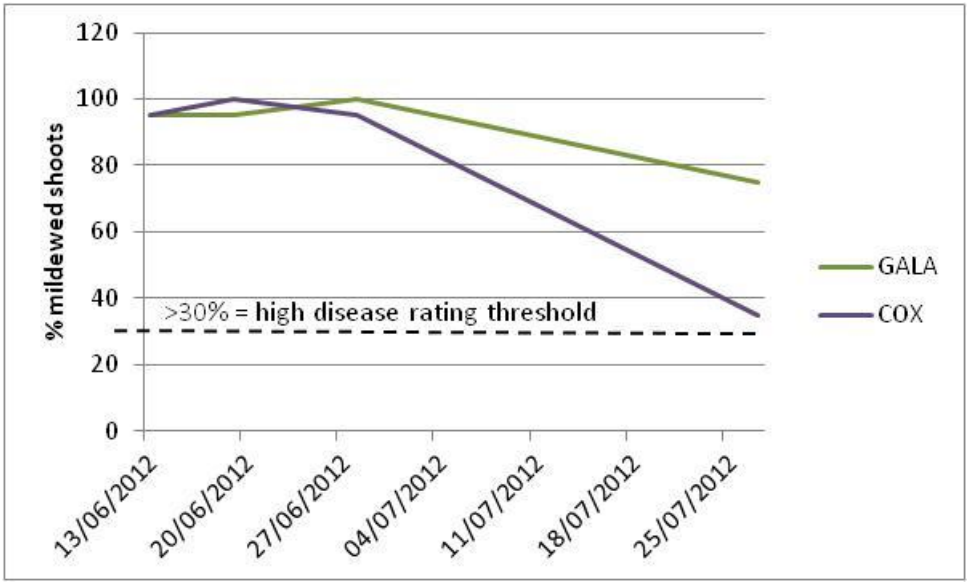


Fig. 3. Mildew monitoring, % mildewed shoots in management plot for cvs. Gala and Cox. Dotted line denotes 30% threshold over which disease rating is considered high.

A total of 11 mildewicide spray applications were made in the management plot (Table 9 and Fig. 4a) with six-eight day intervals compared to seven sprays applied to the routine managed plot with 10-14 day spray intervals. The reduction in the mildew epidemic is evident in the management plot compared to the routine plot in both Cox and Gala (Fig 4 b and c).

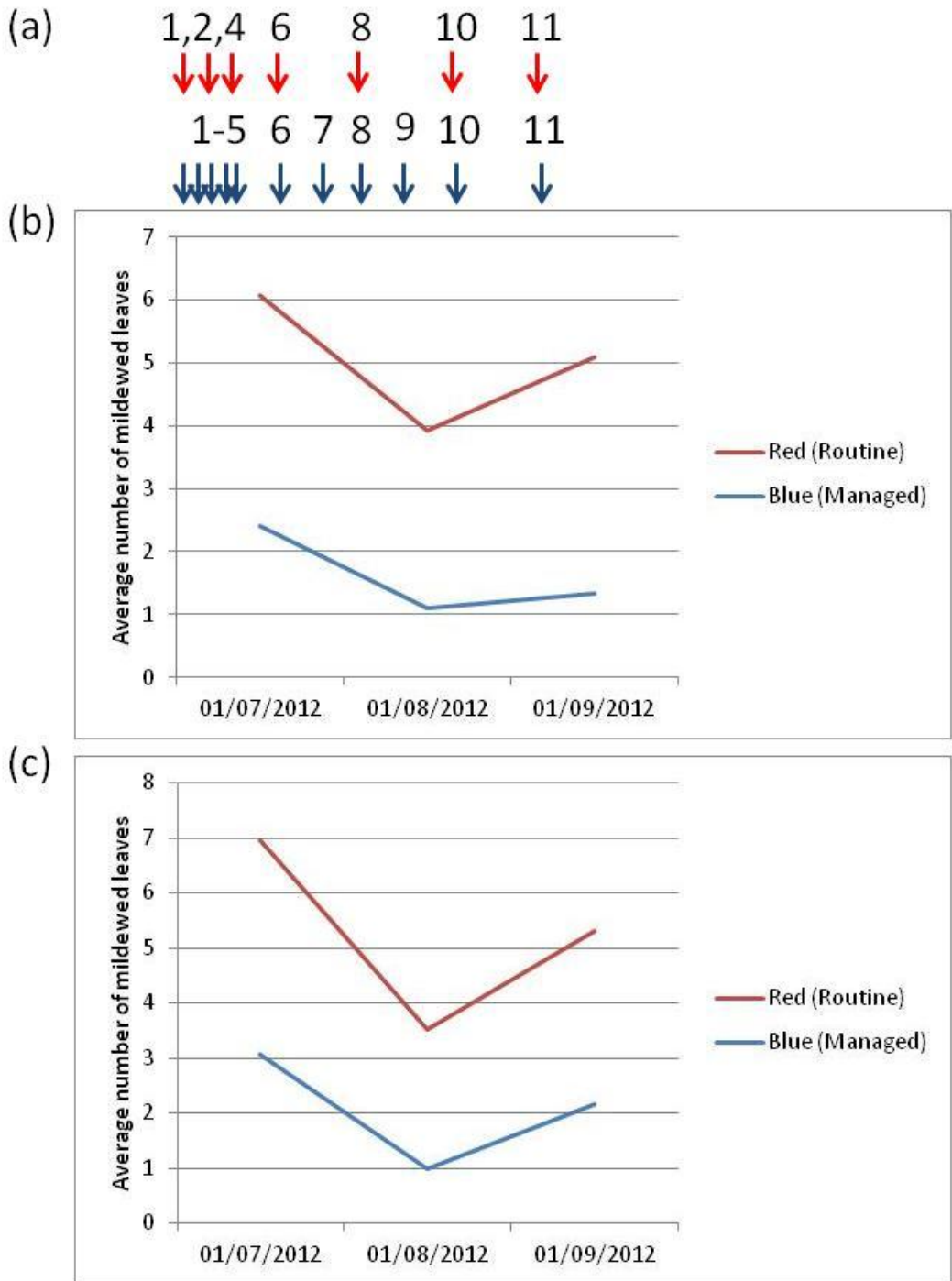


Fig. 4. Secondary mildew epidemic levels and treatments applied for control. (a) treatment numbers applied to routine (red) and managed (blue) plots respectively. Refer to Table 9 for product, dose rate, concentration and specific dates of application. (b) and (c) show average numbers of mildewed leaves in top five leaves of shoot on three assessment dates for Cox and Gala respectively.

Discussion

High levels of mildew inoculum carried over from the previous season and highly favourable conditions for mildew spread made it very challenging to control the mildew epidemic in the trial plots in this single season. As a result of the high level of infection, the managed plot received an intensive programme of mildewicides. The managed plot, unsurprisingly, had a reduced mildew epidemic compared to the routine plot on all three assessment dates. This is the first year of a multi-year trial, and reductions in mildew levels are expected to accrete over successive seasons. Although fungicide inputs are initially higher, it is expected that once the mildew epidemic is under control fewer sprays will be required to achieve equivalent mildew control compared to the routine programme.

In order to fully benefit next year in 2013 from the reduction in the epidemic achieved in the managed plot this year, the orchard will continue to be monitored for shoot extension post-harvest, which mild October weather can sometimes permit. This late extension growth can be colonised by residual mildew, often unchecked by fungicides, which then over-winters in terminal vegetative buds and emerges in the spring as primary mildew. Any late extension growth will trigger a further mildewicide application to the managed plot.

In addition to the mildew management trial commenced this year, an additional trial will be initiated to trial an integrated mildew management programme to include cultural control methods, such as the removal of mildewed tips, along with the inclusion of promising synthetic, alternative and biocontrol mildewicides identified in the apple mildew trial in the CP 77 / HL01109 SCEPTRE project.

Conclusions

- Having started the season with a very high level of inoculum. A reasonable level of control has been achieved through applying an intensive fungicide programme on the managed plot.
- The gains in a reduction in the mildew epidemic in the managed plot are expected to accrete in year 2 and subsequent years.

Knowledge and Technology Transfer

This is the first year of the projects and as such insufficient data has been generated for knowledge and technology transfer at this early stage.

Appendix 1 - Summary of existing projects worked on in year 1

CP 77 / HL01109 SCEPTRE

Evaluating treatments for mildew on apple

- Two trials were conducted to evaluate the efficacy of novel chemical and biological treatments against mildew.
- Several effective new chemical treatments have been identified, biological treatments performed poorly.
- Data will be presented in the SCEPTRE annual report.

Post-harvest treatments for botrytis in pear

- Biological treatments are to be assessed for the control of botrytis in stored pears.
- Treatments have been applied as drenches to crates of fruit seeded with inoculated pears.
- The trial has been set up and will be assessed in March/April 2013.

Evaluating treatments for mucor/rhizopus on strawberry

- A strawberry trial was conducted to assess the efficacy of chemical and biological spray treatments against mucor/rhizopus.
- Several effective new chemical treatments have been identified, biological treatments performed poorly.
- Data will be presented in the SCEPTRE annual report.

Evaluating treatments for crown rot in strawberry

- A strawberry trial was conducted to assess the efficacy of chemical and biological treatments applied as root drenches against *Phytophthora cactorum*, the causative agent of crown rot.
- The trial has been treated and will be assessed twice: in late September 2012 and April 2013.
- Data was presented at the oomycetes workshop organised by the HDC in October 2012.
- The data from this year's trial will be presented in the SCEPTRE annual report.

Hort-LINK projects

HL0191 / SF 94 - Minimizing residues in strawberry

- A commercial trial has been conducted on multiple sites comparing an integrated pest and disease management programme against the growers' standard.
- Assistance has been provided on the day to day disease responsibilities in this trial. An overview of the work was presented at the IOBC conference in Turkey in October 2012.

HL01105 – Blackcurrant biocontrol

- A programmes trial for botrytis control using a combination of chemical and biological treatments to reduce residues on fruit has been undertaken.

- Laboratory work has also been undertaken to elucidate latent infection of botrytis in blackcurrant.
- The results of the former trial were presented at the AAB meeting on biocontrol agents in October 2012.

HL0189 / TF 194 - Plum and cherry biocontrol

- A commercial trial has been conducted on multiple sites comparing an integrated pest and disease management programme against the growers' standard.
- Assistance has been provided on the day to day disease responsibilities in this trial.

TSB

Evaluating novel biofumigation products for effectiveness against *Verticillium dahliae*

- Proficiency has been gained with the Harris plate counting test for assessing levels of *V. Dahlia*.
- Collaboration has occurred with other scientists to evaluate the effectiveness of a novel volatile product.
- Optimised protocols have been developed to assess soil respiration of field trial samples to evaluate whole ecosystem effects.

HDC projects

TF 195 - Apple mildew sensitivity

- Assistance provided in trial set-up and assessment.
- Data will be presented in the HDC annual report.

SF 132 - Blueberry dieback

- A consortium has been assembled to determine the causative agent of branch and bush dieback in blueberry. EMR has the responsibility to undertake field work in this project.
- Crop-walking has been conducted in several blueberry plantations throughout the UK.
- Samples have been collected and sent to FERA for diagnostics.

TF 203 - Mycotoxin production associated with *Fusarium* mouldy core in apple

- This aims to determine whether mycotoxins associated with *Fusarium* are present in infected fruit.
- Isolates have been collected from orchards throughout the UK.
- Molecular identification has determined that *Fusarium* species capable of producing mycotoxins are causing mouldy core in apple.
- Samples are now being collected and prepared for mycotoxin analysis.

Chemical company trials/consultancy activities

- Evaluation of blackcurrant botryticides and chemicals effective against storage rots.
- Thanet Earth – confidential trial, report draft submitted.

Plant clinic

- Guidance in traditional diagnostic skills for >150 plant clinic samples in the 2012 season.
- Introduction of molecular based diagnostic techniques.