Grower Summary

SF 158

Integrated Pest Management (IPM) of Cane Fruit Pests and Diseases

Annual 2017

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GROWER SUMMARY

The overall aim of the project is to advance and optimise on-farm integrated management of key pests and diseases of cane fruit. Within this project, it is planned to work on five differing objectives over the five year duration:

1. Investigate the infection process of *Phytophthora rubi* to inform the use of alternative or supplementary means to the use of chemical plant protection products for reducing the level of root rot in raspberries.

2. Develop and maintain IPM approaches to successfully control two-spotted spider mite whilst controlling spotted winged drosophila (SWD) and capsids with insecticides.

3. Develop and combine novel and current IPM approaches to successfully control blackberry leaf midge;

4. Establish cane management approaches on a model crop to optimise IPM strategies and spray penetration into canopies;

5. Disseminate research results to growers and translate research outputs into practical ‘ready to use’ techniques for immediate uptake on farms.

For ease of reading, this Grower Summary report is split into sections for each of the objectives (pests & diseases) being worked upon. The first year’s work concentrated on Objectives 1 to 4, so only these are reported on in this first annual report.

Raspberry root rot

Objective 1 – Investigate the infection process of Phytophthora rubi to inform the use of alternative or supplementary means to the use of chemical plant protection products for reducing the level of root rot in raspberries

Headline

- Prestop and three coded products showed some promise in controlling *Phytophthora rubi* when compared to Paraat treated plants and an untreated control.

Background and expected deliverables

Phytophthora root rot is now the most destructive disease of raspberries worldwide. Where raspberries have been grown in the soil *Phytophthora rubi* (previously known as *P. fragariae* var. *rubi*) is almost ubiquitous. Outbreaks of this disease across Europe at the same time in traditional raspberry-growing areas suggests that the disease has spread through the propagation network and has been distributed to farms in new planting material (Graham et al. 2011). Current approaches for Phytophthora control rely on fungicide applications twice per year either as a soil-applied drench or through the drip irrigation. SL567A (44.7% w/w metalaxyl-M) and Paraat (500 g/kg dimethomorph) can be used, although resistance developing in pathogens where products have only a single mode of action is a major concern. To reduce the risk of this occurring, agrochemical companies are developing co-formulated products such as Fenomenal (fenamidone and fosetyl-aluminium) which should be gaining approval for cane fruit. Although this will help, none of these types of products can completely control this disease.

The work in this project will focus on understanding the activity of non-conventional products that may improve root health and the production of propagation material that is more resistant to the disease. The current work is being divided into four specific work packages:

Task 1.1: To elucidate whether more-susceptible varieties cause greater attraction of *P. rubi* zoospores than more-resistant varieties.

Task 1.2: To determine if selected elicitors and nutrients have a detrimental effect on *P. rubi* infection.

Task 1.3: To determine if or how selected fungal and bacterial supplements or microbial plant protection products have a detrimental effect on *P. rubi* infection.

Task 1.4: To determine whether non-conventional products (microbial and plant stimulants) have the potential to maintain and potentially improve plant establishment and root health of raspberry plants when applied from propagation onwards.

Summary of the project and main conclusions in year 2

In 2015, Tulameen plants in growing media had been treated with biostimulants and growth promoters following propagation. In 2016, these were potted up at two separate sites - a commercial farm in Oxfordshire and ADAS Boxworth. The plants then received further treatments at each of the sites according to the product labels. Treatments included Prestop (*Gliocladium catenulatum*) as well as products without approval for use in growing media in cane fruit. They were compared with a single dench of Paraat (dimethomorph).

The ability of the products to improve crop performance and vigour was investigated at the Oxfordshire site, but no differences were found between any of the treatments and the untreated control.

At ADAS Boxworth, plants were treated with preventive applications of products before inoculation with *P. rubi*. In some cases, curative applications were also made. No collapse of primocanes occurred during 2016 as a result of root rot, but destructive assessments were made for root rot in early 2017 and the results will be reported later.

Prestop and three of the coded products had slightly, but significantly, increased the number of primocanes compared with the untreated control and Paraat treated plants. No phytotoxicity was caused by any of the products at either site. The Oxfordshire site is planned to continue for a second cropping year with further treatment applications.

Financial benefits

Raspberry root rot (caused by *Phytophthora rubi*) is the most devastating disease currently faced by cane fruit growers and in particular by raspberry producers. The disease spreads rapidly through the root system of the crop, leading to complete death of large areas of a plantation. Where severe, in soil grown crops, it commonly kills 75% of a raspberry plantation within two to three years of establishment. Although perhaps slower to spread in container grown crops, it has a similar effect in killing significantly large areas of a plantation within a few years of planting and establishment. Not only do growers make significant financial losses, they also incur additional labour costs in setting up new replacement plantations more frequently, along with the associated costs of establishing a new plantation along with the support system that goes with it.

Assuming a typical return for raspberries of £6.49/kg to growers (Defra Basic Horticultural Statistics 2014) and a yield of 14 tonnes/ha, then 75% crop loss would lead to a financial loss of £68,166/ha. Increasing the health of propagation material and providing material that is more resistant to the disease would not only significantly reduce such losses but lengthen the life expectancy of a raspberry plantation, thereby reducing the additional costs of re-establishing new plantations on a frequent basis.

Action points for growers

- Consider biological alternatives to plant protection products for the control of *P. rubi.*

Two-spotted spider mite

Objective 2 – To develop and maintain IPM approaches to successfully control two-spotted spider mite whilst controlling spotted wing drosophila (SWD) and capsids with insecticides

Headline

- The use of overhead mist spraying using large droplet size reduces spray coverage on the undersides of leaves in the crop canopy, offering more refuge to predatory mites which control two-spotted spider mite.

Background and expected deliverables

A key current question for growers of soft fruit is how to maintain the successful Integrated Pest Management (IPM) approaches that have been developed over the past 10 years whilst applying crop protection products to control SWD. Two-spotted spider mite (TSSM) can be a devastating pest of raspberries, especially on crops grown under glasshouse or polytunnel protection and during hot weather. Control of TSSM with acaricides requires good spray cover, as most acaricides are contact acting. Effective leaf cover is difficult to achieve in raspberry crops which often have dense canopies. Recent changes in legislation have also meant that there is a limited range of acaricides for use in protected and outdoor raspberries and other cane fruit crops and it is likely that this trend will continue (e.g. abamectin is under threat due to potentially being an endocrine disrupter). The difficulties of applying sprays to a raspberry crop and restrictions on crop protection products mean that predators of TSSM are an important method for the control of this pest.

Phytoseiid predatory mites are the main natural enemies of TSSM. There are two main naturally occurring, overwintering, species in raspberry (predominantly *Amblyseius andersoni* but *Neoseiulus californicus* is also common). These mites naturally regulate TSSM populations to a greater or lesser extent, but not reliably. In recent years, growers have been successfully introducing *Phytoseiulus persimilis* predatory mites and the predatory midge *Feltiella acarisuga* for the control of TSSM mite in outdoor/protected raspberry and blackberry crops. However, information on side effects of crop protection products on biological control agents and experience in other countries, demonstrates that applications of products to control SWD such as spinosad (Tracer), lambda-cyhalothrin (Hallmark) and deltamethrin (e.g. Decis), can adversely affect these biological control agents leading to serious outbreaks of TSSM.

Outbreaks of TSSM and other mites, as a result of disruption to biocontrol by naturally occurring and introduced predatory mites, by sprays of products for SWD and/or capsid bugs, is an immediate serious threat which the UK cane fruit industry faces.

This study aims to address this problem through two specific work packages:

Task 2.1: To investigate consequences of SWD control strategies on two-spotted spider mite populations on a commercial holding already dealing with the pest.
Task 2.2: To develop compatibility strategies for biocontrol of two-spotted spider mite by predatory mites with insecticide sprays for SWD and capsids.

Summary of the project and main conclusions in year 2

In 2015, the effects of overall canopy spraying verses overhead misting application of a programme of sprays of deltamethrin (Decis / Bandu), spinosad (Tracer) and chlorpyrifos (Equity) on TSSM and naturally occurring predatory mites were compared and both the overall effect of date and overall effect of treatment were significant. In early August, the numbers of natural phytoseiid mites were lower in both of the sprayed treatments, possibly following specific spray applications of spinosad. The numbers of TSSM then rose significantly in the sprayed plots from the 17 August 2015. The numbers of SWD were lower in both of the treated plots (spraying and overhead misting).

In 2016, the effects on raspberries in tunnels of overall canopy spraying verses overhead application of a programme of sprays of Decis (deltamethrin) and Tracer (spinosad) on TSSM, naturally occurring predatory mites and introduced *Phytoseiulus persimilis* were compared. Nozzles were used that gave a slightly larger droplet size than in 2015 in order to determine whether this would give less spray on the underside of the leaves, so providing refuge for the predatory mites and therefore improved control of TSSM. The results showed that both treatments reduced the number of SWD compared with the untreated. There was less spray on the underside of the leaves in the overhead spray treatment. TSSM numbers were higher in the sprayed treatments (for all life stages with the knapsack spray). The natural phytoseiids were affected by the spray treatments, but the effect could be mitigated by spraying from above. Introduced *P. persimilis* was less affected by the spray programme than anticipated.

Financial benefits

Before the spotted wing drosophila first arrived on UK shores, raspberry growers had refined their IPM programmes reasonably well and were gaining satisfactory control of two-spotted spider mite using biological and naturally occurring control programmes, primarily through the introduction of the predatory mite *Phytoseiulus persimilis* and sometimes complemented with other predatory midges such as *Feltiella acarisuga*.

The vital importance of controlling spotted wing drosophila at all costs, has resulted in a conflict with IPM programmes, given the nature of the crop protection products used for SWD control and the fact that they upset the predator/prey balance that is developed. However, failing to gain control of two-spotted spider mite can lead to serious reductions in the efficient photosynthetic area of the plant and this can lead to the production of small and shrivelled fruits and a subsequent reduction in the marketable yield of raspberry or other cane fruit crops.

Assuming a typical return for raspberries of £6.49/kg to growers (Defra Basic Horticultural Statistics 2014) and a yield of 14 tonnes/ha, then a 25% crop loss caused by two-spotted spider mite (a typical loss incurred) would lead to a financial loss of £22,722/ha. Developing a refined IPM programme on raspberries which can also cater for the control of other pests such as SWD and common green capsid, will significantly reduce such losses from two-spotted spider mite.

Action points for growers

- Be aware of the contribution of natural predators in the control of two-spotted spider mite.
- Using spray coverage that allows predators to shelter from control products intended for SWD is likely to preserve natural and introduced predators.

Blackberry leaf midge

Objective 3 – Develop and combine novel and current IPM approaches to successfully control blackberry leaf midge

Headline

- *Steinernema kraussei* can reduce blackberry leaf midge emergence from pupae in the ground.

Background and expected deliverables

The blackberry leaf midge (*Dasineura plicatrix)* has become an increasing problem on blackberry, hybrid berry and increasingly raspberry, with double cropping primocane raspberries being particularly vulnerable to attack by this pest. The blackberry leaf midge can have up to four generations per year under protection and causes damage to the leaves and the growing points of plants. This can result in the stunting of cane growth leading to loss of yield. It has been estimated that the midge could reduce raspberry yield by 40% and blackberry yield by 10% (Fountain, 2013). This pest has increasingly been identified as a high priority by the industry, particularly in primocane systems.

Project SF 102 'Biology and integrated control of blackberry leaf midge on blackberry and raspberry' (Bennison, 2011) assessed alternative ground-based methods of control for this pest. Treatments included cultural controls, ground-based predator introductions and entomopathogenic fungi applications to soil/substrate. Promising approaches were identified in laboratory tests, particularly covering the soil surfaces with polythene. Another promising approach included introducing the predatory mite *Macrocheles robustulus*. The entomopathogenic fungus *Beauveria bassiana* (Naturalis-L) was not effective when applied as a soil drench or as a foliar spray.

Since completion of SF 102, another entomopathogenic fungus, *Metarhizium anisopliae* (Met 52) (recommended for vine weevil control) has gained EAMUs for use on rubus hybrids either pre-planting as a growing media-incorporated treatment or as a post-planting mulch for control of midges with a pupal stage in the soil and warrants testing against blackberry leaf midge. A liquid formulation of Met52 (Met52 OD) is now approved in the UK as a foliar spray on various crops including protected strawberry for control of thrips and mites. The product is not currently approved on cane fruit or for use as a drench but it is possible that an EAMU could be sought if shown to be more effective than Met52 granular, as a substrate drench would be more practical than incorporation for soft fruit growers.

Entomopathogenic nematodes were not tested as a ground drench in SF 102 as recent work had been carried out at Wageningen investigating three species of nematodes (*Steinernema feltiae*, *S. carpocapsae* and *Heterorhabditis* sp.) but none were found to be effective against blackberry leaf midge larvae (Wenneker, 2008 and personal communication). *Steinernema kraussei* is widely used for vine weevil control on soft fruit but has not yet been tested against blackberry leaf midge.

Spinosad (Tracer) is used as a drench to brassica modules for control of cabbage root fly and could have potential for control of blackberry leaf midge larvae when they drop to the ground to pupate. Tracer has EAMUs for use as a foliar spray on protected and outdoor crops of raspberry for control of thrips so if it was shown to be effective as a ground drench for control of blackberry leaf midge, an EAMU application could be made for use as a drench.

Project SF 141 'Efficacy of insecticides, timed use of the blackberry leaf midge sex pheromone trap, to control the pest on raspberry' provided a strategy for the control of blackberry leaf midge. However the insecticides used at present are not IPM-compatible and the strategy has practical limitations. Problems with spray coverage and timing mean that other tools are required to limit the threat that this pest poses.

Summary of the project and main conclusions in year 2

The results of a laboratory test in the first year of the project showed that a drench of the entomopathogenic nematode, *Steinernema kraussei* (Nemasys® L) to coir substrate in pots significantly reduced the numbers of adult blackberry leaf midges emerging compared with water controls after adding fully grown midge larvae to the treated substrate surface to mimic them dropping from infested leaf tips to the ground to pupate.

In Year 2, a field trial was done to test drenches of Nemasys® L applied to the soil beneath the crop canopy of a commercial soil-grown raspberry crop under a polythene tunnel. The crop had a history of blackberry leaf midge. Nemasys® L is already widely used by soft fruit growers for control of vine weevil. Two consecutive drenches were made to replicate plots by the host grower, the first on 5 May following the first midge larvae being recorded in leaf tips on 27 April and the second on 6 June following the second generation of midge larvae being recorded in leaf tips on 31 May. Assessments were made of numbers of twisted leaf tips, infested leaf tips and numbers of midge larvae in infested leaf tips every two weeks from 5 May to 4 July.

The mean percentage of twisted and infested leaf tips and mean numbers of midge larvae in infested leaf tips were not reduced in Nemasys ® L-treated plots compared with those in untreated plots on any assessment date. On 5 May when first generation midge larvae were active in leaf tips, there was a mean of 20% and 23% leaf tips infested in untreated and nematode-treated plots respectively. By 4 July, the percentage leaf tips infested had increased to 100% and 93% in untreated and nematode-treated plots respectively.

Possible reasons for the lack of control by Nemasys ® L was insufficient soil moisture to allow nematode movement and survival and the short 'window' of opportunity for nematodes to infest the midge larvae before they spin a protective cocoon in which to pupate.

Financial benefits

The blackberry leaf midge is a relatively new pest of raspberry and blackberry in the UK, having assumed greater importance as increasing crop areas have been protected by temporary polythene tunnel structures in the field. It is not uncommon to find that the midge has reduced raspberry yield by 40% and blackberry yield by 10%.

Assuming a typical return for raspberries of £6.49/kg to growers (Defra Basic Horticultural Statistics 2014) and a yield of 14 tonnes/ha, then a 40% crop loss caused by blackberry leaf midge would lead to a financial loss of £36,355/ha. Developing a novel IPM approach will significantly reduce such losses from blackberry leaf midge.

Action points for growers

- No action points have been developed for blackberry leaf midge at this early stage of the project.

Verticillium wilt

Objective 4 – To investigate strains of Verticillium spp. present in UK cane fruit plantations and the thresholds for infection in blackberry and raspberry

Headline

- Molecular assays for *Verticillium dahliae* allow pathogen presence determination.

Background and expected deliverables

Verticillium dahliae and *Verticillium albo-atrum* are the causal agents of Verticillium wilt in raspberry and blackberry. These fungi have a wide host range of over 300 woody and herbaceous plants. In raspberry and blackberry, the disease can be very destructive resulting in stunted shoots, extensive wilting and ultimately plant death. Crop loss can occur if the canes die before reaching maturity and as plants succumb once established.

Characteristic symptoms in raspberry include: Leaves turning pale and wilting; premature drop of leaves from the bottom up, occasionally leaving only a tuft of leaves at the top of canes. In severe cases, infected primocanes are stunted and develop a blue colour on one side of the cane. Symptoms differ from root rot in that wilting occurs from the top down in root rot, with the characteristic shepherds crook on spawn and suppression of spawn growth.

In blackberry, the infected canes wilt and the leaves turn yellow and become brown and necrotic similar to raspberry. However you do not see the same characteristic blue cane staining.

Severe outbreaks have occurred sporadically in UK cane fruit crops and widespread infection as a lower incidence is suspected. The causal fungi are difficult to isolate from infected canes so diagnosis is often presumptive (i.e. bases on symptoms). Some of the newer raspberry and blackberry varieties being planted by growers are derived from USA breeding lines with known high susceptibility to verticillium wilt.

Verticillium dahliae is considered the primary pathogen and it can survive in the soil for many years. Once susceptible plants are placed in infested ground, the fungus can grow into the xylem and colonise the whole plant. Therefore knowledge of whether the soil is infested with the fungus prior to planting is useful to aid planting decisions.

The expected minimum five year life of raspberry and blackberry plantations can be severely shortened when roots become infected by *Verticillium* species and / or *P. rubi* leading to cane death. Symptoms often do not clearly distinguish between verticillium wilt and phytophthora root rot. The area of primocane-fruiting raspberries has been increasing in the UK and the varieties grown tend to be susceptible to Verticillium wilt. *V. dahliae* and *V. albo-atrum* have a wide host range and persist in many soils. The relative damage caused to raspberry by each

Verticillium species is not known.

The Harris test, a wet sieving method, can be carried out on soil samples before planting to enumerate the microsclerotia of *V. dahliae*. However, many growers do not submit samples because the assay takes 6-8 weeks. *V. albo-atrum* is not detected by the Harris test.

Real-time, or Quantitative, PCR assays (QPCR) for testing soils prior to planting for specific soil-borne *Verticillium* species using DNA extracted from large volumes (up to 1 kg) of soil have now been successfully developed (project SF 97). The techniques were initially developed during Potato Council-funded potato diagnostics research (Project R253) and utilize pre-extraction processing, buffers to remove reaction inhibitors and an automated DNA binding system to capture total DNA. These tests provide results within a few days (rather than 6-8 weeks for *V. dahliae* microsclerotia). PCR detects DNA in both dead and live cells, but microbe DNA deteriorates quickly in normal conditions and so fungicide/fumigant killed pathogens would rarely be detected. Detection of *V. dahliae* using QPCR has been achieved down to levels correlating with 0.5 microsclerotia / g soil and the assay is still being improved in order to attain a detection of <0.5 microsclerotia / g soil. It is already possible to detect below one microsclerotia by testing multiple soil extractions, but this increases the cost of the test. Low detection rates may not, however, be as important for raspberries as it is for strawberries. Strawberry growers benefit from information on the relative susceptibility of strawberry varieties to certain levels of *V. dahliae* microsclerotia (e.g. 5-9 microsclerotia / g soil can cause 50% loss in variety x) and can base their variety selection on this information. However, the threshold for damage in *Rubus* is not known. Observations suggest a tolerance of up to 50 propagules / g soil for some commercial floricane raspberry cultivars, while some primocane fruiting cultivars may be ten times more susceptible.

Current *Rubus* varieties come from a number of breeding lines and so differences in susceptibility to *Verticillium* and *Phytophthora* are likely. The James Hutton Institute has a programme of screening lines for *Phytophthora* resistance using molecular markers. Two regions of the genome influence susceptibility to the disease. Molecular markers to aid *Verticillium* resistance breeding have not been found although variable resistance between *Rubus* species has been noted (with black raspberry very susceptible) and so there is potential for variety rankings and the production of thresholds for V*erticillium* spp. in the soil for different varieties.

V. dahliae is also a very diverse fungus with different strains reported that may differ in pathogenicity. Relatively little is known about verticillium in UK blackberry and raspberry crops. Nothing is known about which strains are present, their pathogenicity to individual species, the importance of soil-borne inoculum, thresholds for causing disease, and disease development in container grown crops. Work on verticillium wilt in strawberry in Project SF 97a resulted in the development of a molecular 'tool box' of real-time PCR assays for the detection of various verticillium strains directly in soil and also showed there was some diversity in the UK of *V. dahliae* strains present.

In this project, these new molecular tools will be used to determine which strains of *V. dahliae* are present in UK cane fruit crops and work towards determining the threshold for causing disease in raspberry and blackberry.

Summary of the project and main conclusions in years 1 and 2

In Year 1, *Verticillium dahliae* was detected in raspberry and blackberry plant material from the field but was not detected in young plants from three propagators. There was a relationship between DNA of *V. dahliae* in the stems and in the roots although DNA of the pathogen in stem bases was considerably higher. This will inform future sampling strategies. Little *V. dahliae* DNA was recovered from the soil under plants with *V. dahliae* DNA and the reasons are unclear.

A range of fungi were isolated from the plants including Fusarium, Alternaria and Pythium. Interestingly, Ilyonectria species were isolated from two plants and this was confirmed by DNA sequencing of the rDNA region. Ilyonectria is a known pathogen of raspberry and may also be causing plant death. Further work in this project will attempt to characterise these isolates further by sequencing additional loci.

A novel sequence from the *V. albo-atrum* EF region has been identified and four TaqMan assays have been designed and tested for specificity. Although the TaqMan assays detected *V. albo-atrum* and not *V. dahliae*, they did cross react with *V. tricorpus*. An alternative assay site has been identified. To validate the VCG1 assay, isolates belonging to different VCGs are in the process of being collected. IGS primers suitable for determining VCG through IGS sequencing have been identified and will be used once this isolate collection is complete.

Further work conducted by Fera validated a DD assay demonstrating that it is able to detect *V. dahliae* and *V. longisporum* to an acceptable level. The assay could therefore be used to detect infection of plant material by these fungi and assess infection levels in soils. The assay was used in 2015 for the survey of raspberry and blackberry plants, planting material and soils.

Financial benefits

Verticillium wilt of raspberry and blackberry has become a much greater threat to raspberry and blackberry growers in the past 15 years. Many of the modern primocane raspberry and the recently introduced blackberry cultivars are particularly susceptible to *Verticillium dahliae* (the cause of Verticillium wilt). Since it began to cause crop loss, plant pathologists and cane fruit growers have been lacking in the knowledge of how susceptible different cultivars are to the disease at differing levels of the pathogen in field soils. This has made it difficult to make management decisions about the safety of a new field soil which has never before hosted a cane fruit crop.

By improving our knowledge of this and developing threshold levels of soil inhabiting Verticillium dahliae for different cultivars, it will allow growers to make informed decisions about the safety of a new field soil which might be used to establish a new crop, thereby avoiding severe crop losses to the disease in the first two to three years of a plantation's life span.

Action points for growers

- Note that stem necrosis is a better determinant of the presence of Verticillium causing wilt than leaf wilting alone.