

Project title: Identifying novel powdery mildew susceptibility/resistance genes in strawberry. Project number: SF/TF 170: CTP PhD Studentship Scheme **Project leader:** Dr Helen Cockerton Report: Annual report 2020 **Previous report:** N/A Key staff: Professor Jim Dunwell Location of project: **NIAB EMR** Harriet Dunclafe **Industry Representative:**

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



AUTHENTICATION

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

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GROWER SUMMARY

Headline

Analysing strawberry powdery mildew (*Podosphaera aphanis*) to identify key mildew resistance/susceptibility genes using a Genome Wide Association Study (GWAS).

Background

Strawberry is a major soft fruit produced in the UK, favored for the sweet fragrant taste and containing compounds with potential health benefits (Amil-Ruiz *et al.*, 2011). The introduction of tunnels benefited strawberry crops through protection from extreme weather conditions, extending seasonal production and improving harvest yield. However in these conditions diseases such as powdery mildew can be more prevalent in warm and humid environments (Asalf *et al.*, 2013; Hall, Jin and Dodgson, 2019).

Podosphaera aphanis is a global pathogen, causing disease on strawberry. The symptoms of the disease include upward curling of the leaf edges and discolored patches as well as powdery white mycelium growth which can cover the leaf, flowers and fruit. Infection of the fungus can impact on strawberry development causing misshapen and damaged fruit leading to severe yield loss due to unmarketable fruit (Hall and Jin, 2017; Hall, Jin and Dodgson, 2019).

Powdery mildew is mitigated by the application of stringent crop management practices and fungicide sprays. The fungicide sprays are applied at regular intervals using chemicals such as myclobutanyl or demethylation-inhibiting fungicide (DMI) (Carisse and Bouchard, 2010). However, the frequency of fungicide application can be restricted, as some products such as Brupirmate, Penconazole or Proquinazid can only be applied a few times a year. These restricted applications reduce the possibility of insensitivity and fungicide resistance (Hall, Jin and Dodgson, 2019). For instance, sterol demethylation inhibitors (DMIs) fungicides like Penconazole are highly effective at controlling the disease and is commonly used in commercial fields. However, over time the powdery mildew fungus has developed resistance to these fungicides, current evidence suggests resistance is caused by to mutations in the CYP51 gene (Sombardier *et al.*, 2010).

Summary

The first year of this PhD studentship has required the propagation of 350 different strawberry cultivars. Five replicate plants of each cultivar were propagated and transferred into the field. The process involved the collection of runner's plants, producing misted tips and planting. Genotypic data will be established through DNA extractions from each cultivar and genotyping on the istraw35 Affymetrix SNP chip. Whilst phenotypic data will be collected in year 2 and 3 through disease symptom scoring and imaging of leaves and fruit. The genotypic and phenotypic data will be used in a Genome Wide Association Study (GWAS). The GWAS will identify key Quantitative Trait Loci (QTLs) associated with disease resistance. Disease resistance in plants can be controlled by either a single gene or multiple alleles which are frequently identified through association with QTLs (Pu et al., 2020). The QTLs are represented by molecular markers that correlate with the disease phenotype. Single nucleotide polymorphisms (SNPs) molecular markers are used due to their high frequency in the genome, ease of identification and cost-effective nature (Zhu et al., 2008).

As part of a literature search, key genes associated with resistance to powdery mildew were identified. Bioinformatics tools were used to establish evolutionary conservation of genes against available strawberry genomes from GDR (Jung *et al.*, 2014) - octoploid *Fragaria* x *ananassa* 'Camarosa' and diploid *Fragaria vesca*. These genes were concluded to have a high evolutionary conservation within the subspecies strawberry genome and therefore good candidates for gene editing to validate gene function. Initial stages for gene editing has involved sub culturing of the strawberry cultivars 'Hapil' for is known susceptibility to powdery mildew and 'Calypso' known for its high transformation success. In addition to the bioinformatic design of primers for vector construction.

Financial Benefits

No recommendations at this early stage

Action Points

No action points are available at this time as this is early stages of the project