



Grower Summary

**Systemic infection and symptom
expression of *Neonectria ditissima*
in relation to endophytes conditioned
by environmental stresses**

TF 226

Project title: Systemic infection and symptom expression of *Neonectria ditissima* in relation to endophytes conditioned by environmental stresses

Project number: TF226

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Location of project: East Malling, and several farms in Kent

Industry Representative: Peter Checkley

Date project commenced: 1 October 2017

Date project completed
(or expected completion date): 30 September 2021

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Most of 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.

GROWER SUMMARY

Headline

- One fungal endophyte showed good biocontrol potential against infection of leaf scars by the canker pathogen.

Background and expected deliverables

European Canker, caused by *Neonectria ditissima*, has become the most damaging disease of apple in recent years across all major apple growing regions worldwide. Modern cultivars lack effective resistance to this pathogen and in Europe, the most efficacious methods of chemical control are no longer available. Cultivars differ in their susceptibility but there is no absolute resistance. Previous work, conducted at NIAB EMR, has demonstrated that asymptomatic infection in nursery trees is a significant source of the disease in production orchards. The most economically important damage occurs when the nursery-borne latent infection becomes active and develops into canker on the main trunk during orchard establishment (within three years of planting) – leading to tree death. Ample empirical evidence suggests that stresses following planting can promote symptom expression of those nursery-borne latent infections.

An endophyte is a microbe that lives within a plant for at least part of its life cycle without causing apparent disease. Endophytes have been found in all species of plants studied to date although the endophyte/plant relationships are not well understood. Certain microbial endophytes can help plants to tolerate biotic stress, such as attacks by plant pathogens and herbivory, or abiotic stresses, including salt, drought, or heat stresses. It has been shown in numerous host species that recruitment of specific microbes into the rhizosphere is partially under host genetic control and there is increasing evidence that host genetics influence the microbes occupying the endophytic niche. Endophyte composition can also be influenced by pathogen presence and crop management practices. Current research focuses on how we could exploit endophytes to produce crops that grow faster and are more resistant and hardier than crops lacking specific endophytes.

Recently, we have obtained preliminary data showing a link between antagonist fungal endophytes and cultivar tolerance to *N. ditissima*. One fungal endophyte group, identified as belonging to the genus *Epicoccum* (most likely as *E. purpurascens*, previously known as *E. nigrum*), is much more abundant in two canker-tolerant cultivars than in two canker susceptible cultivars. *Epicoccum purpurascens* is a known antagonist against *Monilinia laxa* (causing stone fruit brown rot) and is being commercially exploited for control of brown rot on stone fruit. It is natural, therefore, to speculate whether the abundance of *E. purpurascens* is related to tolerance to canker development and, if so, whether we could exploit *E. purpurascens* for canker management.

In this LINK project, we aim to build on the preliminary data to investigate whether cultivar differences in tolerance to *N. ditissima* are associated with specific endophytes and, if so, identify the organism(s) and conduct further *in vitro* and *in vivo* biocontrol assays to assess specific endophytes against *N. ditissima*. As well as the direct effect against the canker pathogen we shall study whether these specific endophytes could reduce canker development by inducing host defence systems against the pathogen. To improve breeding for canker resistance, we shall determine to what extent the recruitment of specific endophytes is genetically controlled by hosts by mapping QTLs (quantitative trait loci) and to determine the

extent of overlaps of these QTLs with those mapped for canker resistance. We are conducting experiments to assess (1) to what extent recruitment of endophytes is influenced by soil characteristics and host genotypes, and (2) whether canker symptom expression is related to planting times or the abundance of specific endophytes across several orchards. Finally, to assist in canker management, we are investigating the extent to which endophyte profiles of a specific apple genotype can be influenced by management practices (irrigation and soil amendment).

Summary of the project and main conclusions in Year 3

We have successfully initiated all experimental studies on time; however, much of the lab molecular work has been delayed by at least six months because of COVID-19. We have applied for 6 month no-cost extension and are waiting for response from BBSRC.

- (1) We have profiled endophytes at leaf scars of eight cultivars with differing tolerance/resistance to apple canker:
 - a. Endophyte diversity was primarily affected by orchard location, followed by the scion, whereas the effect of rootstock was small.
 - b. Several fungal and bacterial groups had differential relative abundance between canker resistant (tolerant) and susceptible cultivars. The specific fungal groups included fungal antagonists as well as plant pathogens.
- (2) One *Eppicocum* endophyte from apple has been shown to have good antagonistic effects against the apple canker pathogen in field tests:
 - a. Co-inoculation of both *Eppicocum* and canker inoculum at leaf scars can reduce the canker incidence at leaf scar by 50%.
 - b. For pruning cuts, there is very limited effect of *Eppicocum* probably because of greater susceptibility of fresh pruning cuts combined with a high dose of pathogen inoculum applied.
- (3) We conducted experiments to investigate how quickly and how far *Epicoccum* can colonise apple shoots through inoculated leaf scars. The results will be obtained by early 2021 (COVID-19 permits).
- (4) Inoculation of plants with Plant Growth Promoting Rhizobacteria or Arbuscular Mycorrhizal Fungi at the planting time appeared to result in increased tree development but have negligible effects on canker development.
- (5) Longer duration of trees in cold storage initially led to increased canker incidence post-planting but two years after planting canker incidences did not differ much between the two planting (storage) times.

Financial benefits

The results are from only the second year of a four-year project and hence it is too early to quantify benefits to growers. However, the result that impacts commercial apple production most is the effect of storage duration on canker development.

Action points for growers

- At this stage of the project, there is only one action to recommend to growers: plant trees as soon as possible after lifting.