Healthy trees, healthy future
Enabling technologies to combat *Phytophthora* diseases

News

SCION

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HTHF - Celebrating six years of success

New Zealand is now better able to respond to the threat of the *Phytophthora* pathogens that are affecting trees in horticulture, forest plantations and natural ecosystems thanks to the protocols, baseline data, inoculation systems, analysis pipelines, modelling capacity and germplasm resources established by our HTHF team.

We have met (and exceeded) the programme's overall objectives of determining the susceptibility and biosecurity threat of a wide range of significant *Phytophthora* species to New Zealand's forestry, agriculture and natural ecosystems while identifying pathogen resistance in each host. We have pioneered the proactive genus-wide approach to disease breeding, management and research, establishing New Zealand as world leaders in *Phytophthora* research. Our improved understanding of these pathogens has put New Zealand in a far stronger position to protect our export markets and natural resources against existing and future pathogen threats. We have made considerable inroads in identifying commonality between the disease mechanisms of *Phytophthora* species moving toward more prescriptive approaches to manage future, established and emerging diseases.

Work identifying resistance in kauri is progressing on several fronts with the screening of kauri lines at Manaaki Whenua-Landcare Research consolidating earlier observations of genetic variation in the response of kauri to infection by *Phytophthora agathidicida*. Building on earlier work that targeted seed collection from large, healthy kauri trees within sites that have had long-term exposure to *P. agathidicida*, this year the HTHF program has led the establishment of a kauri germplasm archive resulting in the collection of kauri seed from 650 mature trees from across the range of kauri. Working with 14 mana whenua groups from Kaitaia to Tauranga, we established a significant resource for future screening and kauri research. Work is ongoing to identify genetic markers for resistance to infection with samples submitted for genotype analysis.

The model of Māori engagement within the HTHF programme is being increasingly recognised by both the research partners of the HTHF programme and the mana whenua partners as being a new way of working in biosecurity and specifically within the kauri dieback space.

Our epidemiological and impact studies for red needle cast in radiata pine have set the ground-work for improved management of the disease by foresters. Elucidating the key drivers and environmental limitations of the disease is leading to temporal and spatial disease risk models which are fundamental to targeting the deployment of resistant germplasm and timing of chemical control in the standing forest estate. Work within the HTHF programme directly informs applied research underway between Scion and Forest Growers to establish an integrated pest management approach for red needle cast and *Phytophthora* needle blight.

Screening of apple lines produced from a controlled cross between two selected lines screened earlier in the programme has enabled Plant and Food Research to progress to field trials to further confirm resistance to *P. cactorum* in apple rootstocks. Results from this research have been provided to the ongoing apple rootstock breeding programme at Plant and Food Research with the intention of identifying further markers for resistance to accelerate *Phytophthora* resistance screening.

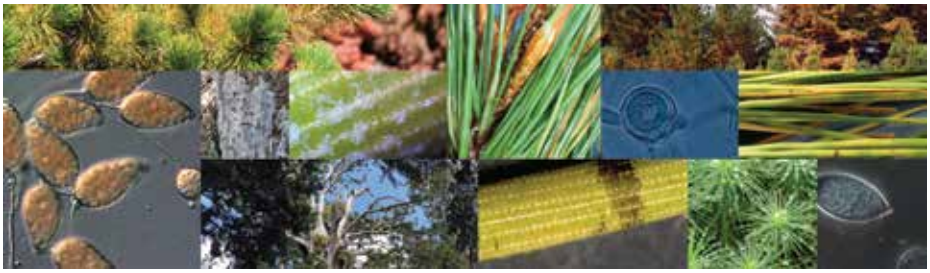
The progress made through this programme on solving *Phytophthora* issues has been outstanding. It is clear that the aspects of this programme that need to continue will do so. Scion and the forest growers are developing a radiata pine research programme that will transition our HTHF programme to one that will help make our pine plantations more resilient to biosecurity threats. As described above, Plant and Food will continue work on identifying apple cultivars resistant to *Phytophthora*. The kauri research will continue and expand from determining in the laboratory if resistance in kauri exists, to actually testing that in the field with mana whenua kaitiaki. We also hope to look at the diversity of kauri over the landscape and determine drivers for seed production and fertility. We foresee a bright future for pine, apples and kauri made possible from the foundation this programme has laid down.

The science is undeniably impressive, but the relationships formed during this journey will also be an enduring legacy and be the foundation to our work as we continue post HTHF.

Thanks to everyone involved in any way in this programme – it wouldn't have been nearly what it was without our collaborators (national and international), students, Programme Overview Committee and Advisory Groups, review team, visiting scientists and the Scion team.

The rest of this final newsletter, is made up of reflections from a few team members across the programme.

Enjoy
Nari Williams and the team



Natalie Graham - Scion

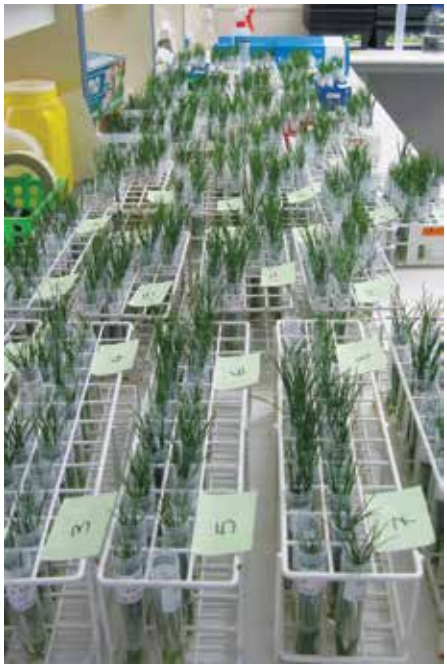
Needles – hundred of thousands of needles! Hand-picked over several months in the Scion nursery under conditions ranging from blazing summer sun to relentless rain or finger-numbing frosts. If the Forest Protection lab magicians could coax the *Phytophthora* into producing sufficient spores, then the needles were saved from premature demise in the bin and we rejoiced that our efforts weren't in vain!

Trays and trays of needles.

The needles were inoculated with *Phytophthora pluvialis*, then carefully laid out on trays (woe betide those who dropped them!) and we waited for biology to do it's thing. Every single needle was then individually eye-balled and red needle cast lesions painstakingly counted and measured by an amazing dedicated team of people, all so we could try to quantify and rank resistance to this disease.

Interpreting the massive amounts of data took almost as long as collecting it. But we persevered and the HTHF programme was able to generate the first set of estimated breeding values for red needle cast resistance in an elite Radiata Pine Breeding Company population, using the detached needle assay.

I am tremendously proud of the effort that this project took. The next challenge is expanding to field-based observations.



Needle testing in the lab.

Stan Bellgard

Systems biology delivering enabling technologies

The sterling, inclusive leadership of Scion and its research partners have advanced the knowledge of *Phytophthora* species and increased the capacity of the collaborating scientists and technologists.
Through the HTHF program we have:

- Developed a *Fluorescent in situ hybridisation* assay using the PCR-primer specific for *P. agathidicida* to assist with visualisation of infection.
- Characterised the *pathology and disease expression* of kauri dieback and deliverd a standard operating protocol for soil and tree-based detection, completing some of the research commenced by the late R. Beever and Ian Horner in 2006.
- Characterised the early *infection and mechanisms of tolerance* and searched for disease tolerance in the kauri populations in partnership with mana whenua of kaurilands, and Scion.
- Characterised the *mycorrhizae and endophytes of kauri root nodules* and studied them using NGS as well as conventional mycorrhizal clearing and staining techniques.
- Complimentary studies have revealed that there are culturable endophytic dark septate endophytes that are directly antagonistic against *P. agathidicida*.
- Engaged tamariki and the next generation of *Phytophthora* biologists through *Unlocking Curious Minds*.

Beyond the science endeavour there is the science translation and cross-cultural relationship-building which will endure long after the HTHF programme finishes. The team have had the opportunity to share knowledge with Māori kaitiaki, and this has served to enrich the experience and provides for an exemplar of "trans-disciplinary" empirical and social science endeavour.

We have learned that knowledge is not held in any one domain or institution, and that it is only through an open sharing of knowledge between participants that we can all move together towards a long-term, integrated solution to "Keep Kauri Standing".

Stefan Hill and Laura Raymond - Scion Metabolomics team

Plants are amazing organisms able to convert sunlight, water, carbon dioxide into more plants! However, this simple description hides the fact that a plant consists of hundreds of thousands of chemicals.

Metabolomics is a hunt in the multitude of chemicals for those that are indicative of infection and in our case - specific responses to *Phytophthora* attack. And just to make the task more challenging, infection of a plant means we are looking at the combined metabolomes of the host and the pathogen.

Using some of the most advanced analytical instruments on the planet, we were able to show many chemicals were related changes in the plants in response to infection. Once identified, these chemicals were mapped to the massively complicated plant metabolomic pathways (the chemical map of life itself).

Laura and Stefan preparing kauri.

Over the last five years, Laura, Ilena, Stefan and Keryn have had opportunities to collaborate with Universities, other CRIs, and Māori organisations. This has led to continuing scientific interactions beyond HTHF and more importantly the development of enduring friendships.

Ngarimu Besselink - Patuharakeke

Ngarimu checking the kauri cones and seed.

In March I went to Pukekauri as part of our Patuharakeke Kaitiaki team to help collect kauri cones as part of the establishing whakapapa lines project for screening for resistance to Kauri Dieback.

I got to join Freddy Hjelm (BioSense Ltd) and the other arborists and climb one of our rangatira kauri trees on the slopes of our pa site, Rangiora. Once I got in the harness I abseiled up a massively tall kauri tree into the canopy. I could see right out over the Whangarei Harbour, it was awesome.

Our trees had heaps of cones that were sent down to Scion in Rotorua for screening. I decided I wanted to see what happened to our kauri seeds when they got to Scion. In May I visited Scion and Gordon gave me a tour of the nursery where I could see our baby trees growing. I went and saw the freezer where the seeds are stored before they are planted out. I also went to the lab where I looked through a microscope and saw what *Phytophthora agathadicida* looks like.

I decided I wanted to do my science fair project for school on kauri dieback. I am going to look at whether kauri dieback infected soil affects seed germination. I will use three trays; one with clean soil (from Pukekauri which is kauri dieback free), a control soil (potting mix) and infected soil (from a confirmed infected site somewhere else in our rohe) and compare them. I think I would also like to know if our rangatira seeds make any difference so I will do the same trial using only the seeds from the tree which I collected from. I would like to thank Scion for showing me around, it was really interesting.

Ngarimu in the kauri house.

Colin Faulds - Scion

It's been a privilege to be the caretaker of the thousands of kauri seed that have come from all over Northland, Auckland, Coromandel and Tauranga during the past four years. I know the huge effort taken by a lot of people to collect these treasured seed and so I have made sure they have had the best possible growing management and conditions possible.

The newly established *Phytophthora*-managed kauri house has grown annually to meet the requirements of each growing phase, which has been appreciated by both me and the team, and the seedlings.

It is great to now be looking at the repatriation of the extra kauri – those not required for testing – to their home rohe. This programme has been a memorable opportunity to be a part of the conservation of this treasured species.

Colin pricking out kauri seedlings.

Chantal Probst – Manaaki Whenua Landcare Research

Chantal (right) carrying out testing in the lab.

Finding the best method to test kauri for tolerance to *Phytophthora agathidicida* proved to be somewhat challenging. We started off inoculating detached leaves and shoots but soon moved on to inoculating small seedlings in petri dishes which died in a matter of days. We finally decided to use 18-year-old seedlings. Kauri seeds from hundreds of trees have been collected over the past four years. We quickly realised that it was near impossible to reach kauri cones with the mere use of tree loppers and we soon found ourselves surrounded by a group of tree climbers. We had to carry heavy loads of cones and soil samples in our backpacks and make sure we would not be spreading this pathogen along the way, but the breath-taking scenery was worthwhile. All the cones were sent to Scion where they were dried, and seeds were sown or stored. Thousands of seedlings have since been grown at Scion and some have been sent to Manaaki Whenua – Landcare Research where they have been inoculated in the hope that we will find *P. agathidicida* resistant kauri families. After inoculation, it is a waiting game until the seedlings die and tissue pieces are plated onto selective media to recover the pathogen. Overall, from collecting kauri cones out in the field to looking at samples under the microscope, the HTHF has been an incredible journey. It has enabled us to meet so many diverse people who have bonded together for their love of kauri and their willingness to preserve this majestic tree.

To learn more about this programme

Contact Dr Nari Williams at nari.williams@scionresearch.com
Visit our website www.healthytrees.co.nz

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