RESEARCH UPDATE

A Summary Of Research 2010

Forest Biosecurity Research Council

Members of the Forest Biosecurity Research Council:

NZ Forest Owners' Association Inc., Scion, Bio-Protection, Ministry of Agriculture and Forestry, Forest Health Research Collaborative, Radiata Pine Breeding Company Ltd.

Members of the Forest Biosecurity Research Council:

Acknowledging the generous support of FRST

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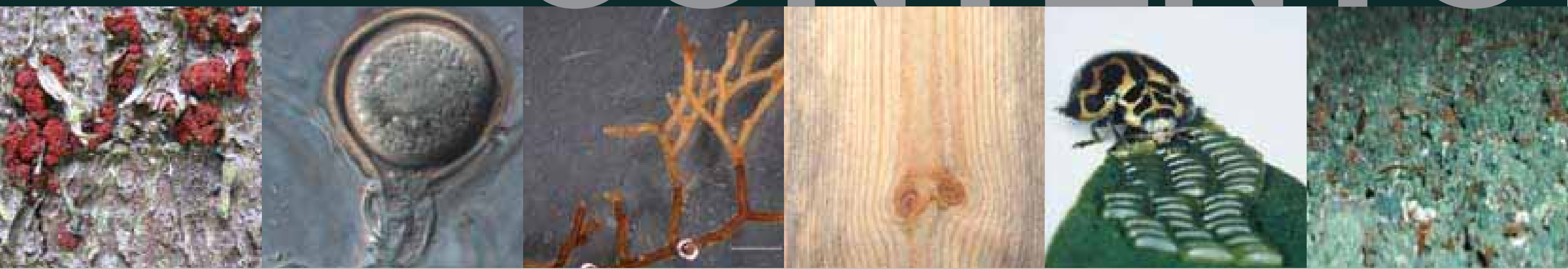

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FOREWORD

DAVID BALFOUR (CHAIRMAN)

The past year has seen a number of forest health scares internationally that reinforce the importance of biosecurity for New Zealand

Larch plantations in Britain have come under attack by an unpredictable fungal pathogen, *Phytophthora ramorum*, causing forest managers to undertake widespread felling of affected areas. Closer to home, Australian foresters are grappling with the recent arrival of guava rust, *Puccinia psidii*, which is expected to cause severe disease of eucalypt species in nurseries and pose a risk to native flora.

While damaging invasions of forest pathogens and pests are nothing new, these events seem to be increasing in frequency overseas. Here in New Zealand we are not immune to such incursions. Foresters watching the upheaval caused by a foreign pathogen in the kiwifruit industry are reminded of the vulnerability of our own industry. We can therefore take heart from achievements made in managing the forest pests and diseases that have already taken hold.

Over the past year we have reached a real milestone in the management of Nectria flute canker. Foresters in southern plantations are now confident that they have the tools and knowledge to minimise the impact of this unexpected disease in radiata pine (see page 5). At the same time, new diagnostic methods

"Over the past year we have reached a real milestone in the management of Nectria flute canker."

have been developed for a range of fungal pathogens that will help to minimise the risk of new incursions by known threats (see page 9). Other research summarised in this report is aimed at finding solutions to problems identified by forest growers as detrimental to the industry.

The projects described in these pages represent a fraction of the total biosecurity research needed by our industry. The serious nature of new and emerging risks has driven a review of biosecurity research priorities and the way the science is managed and funded in New Zealand. Both Scion and the New Zealand Forest Owners' Association surveyed forest growers and other forest biosecurity stakeholders and received clear messages that changes are needed to meet new challenges and priorities.

The Government's imperative to make better use of research capability for the economic benefit of New Zealand has resulted in clarification of the core purpose for each of the Crown Research Institutes (CRIs). As the forestry-focussed CRI, it was pleasing to see that Scion is the lead CRI for forest biosecurity and risk management. As forest managers and owners, and administrators of national biosecurity systems, we are also compelled to become ever more engaged in forest biosecurity research. The FBRC has proven to be an important vehicle for this engagement to occur.

I would thank all of the FBRC members who continue to work hard to ensure maximum benefit is gained from the research investment and that the fruits of this investment are applied to protecting our collective resource.

FBRC PROJECTS

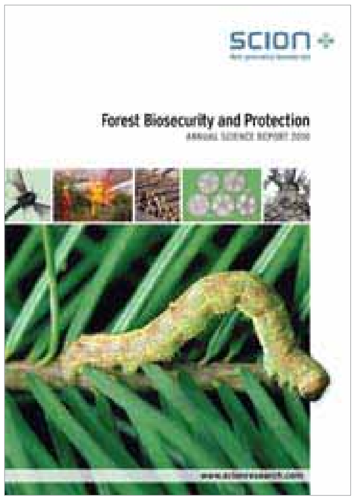
This section of the report summarises projects co-funded by the FBRC.
A brief summary of the project titles and funding contributors is given below.
The contents of this report cover the 2010 calendar year.

ALLOCATION OF EFFORT SPENT ON EACH OF THE FBRC PROJECTS ($000)

Project	FBRC	FRST	Industry	Total
Understanding Nectria	140	151	18	309
Rapid identification	15	102		117
Ectomycorrhizal fungi	20		7	27
Windthrow and degrade from sapstain	25	94	20	139
Biological control of gumleaf skeletoniser	10		51	61
Re-establishing Cleobora	10	45		55
Microbial control of pests and diseases	50	330	124	504
Total	**270**	**722**	**220**	**1212**

GOVERNMENT SUPPORT FOR FOREST BIOSECURITY RESEARCH

The industry funded FBRC projects described in this report are underpinned by substantial Government support from the Foundation for Research, Science and Technology (FRST). This additional funding provides benefit to the forest industry through comprehensive research programmes, the largest of which is delivered by Scion.



The Scion Forest Protection team covers the entire risk management spectrum including:

- Excluding new pests
- Managing existing pests
- Protecting trade of forest products
- Rural fire research

For a full summary of these programmes see the Scion Forest Biosecurity and Protection Annual Science Report 2010 – available through Scion.

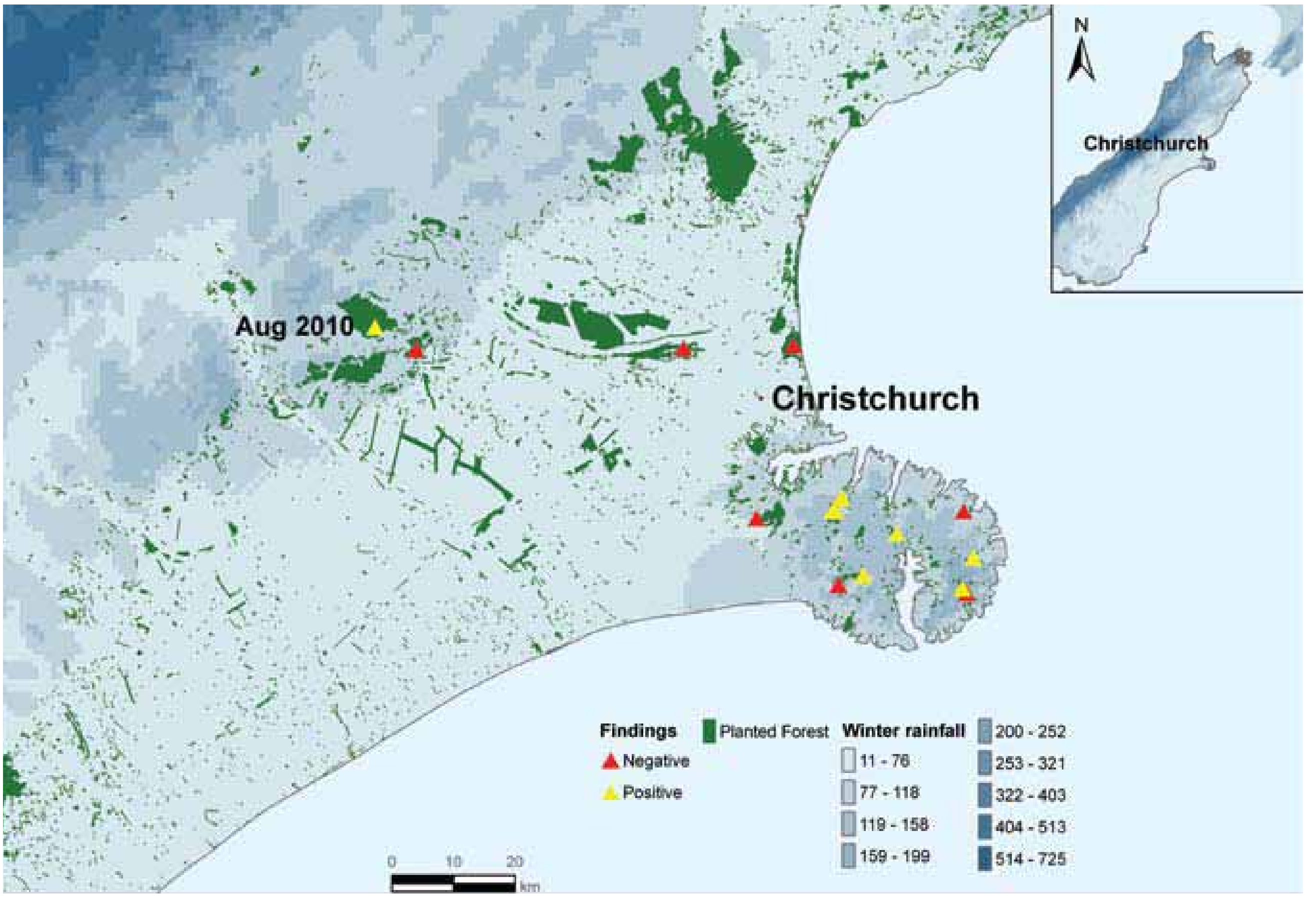
Contact Rose.O'Brien@scionresearch.com for a copy

Through improved knowledge of Nectria flute canker, forest growers have been able to adapt management practices to reduce infection risk

Neonectria fuckeliana is the causal agent of Nectria flute canker, a stem disease of *Pinus radiata* currently occurring only in New Zealand and Chile. The fungus is native to the northern hemisphere, to Europe and North America although it is not an important pathogen within its native range. The disease is found in the southern half of the South Island.

Nectria spread in mid Canterbury

Studies have been carried out in mid Canterbury, at the known limits of Nectria flute canker distribution to determine how far it has spread. In 2009, *Neonectria fuckeliana* was isolated from trees at seven out of 11 sites examined on the Banks Peninsula, and at one site sampled



at Geraldine in South Canterbury. This result confirmed the presence of the Nectria flute canker at those sites and included four new forest location records on the Banks Peninsula for the pathogen.

In early 2010, *N. fuckeliana* was again isolated from Geraldine and from the Banks Peninsula. Intensive sampling and assessment of the three 100-tree plots situated on Banks Peninsula indicated that fluting and infection was increasing in trees pruned within two years, but was decreasing in trees pruned five to six years ago. This observation agrees with results from the pruned stub trial established in Otago on which disease management protocols are based.

Neither Nectria flute canker nor *N. fuckeliana* were found on any of the three sites studied on the Canterbury Plains, despite their proximity to Banks Peninsula. However, in August 2010 Scion field crews found Nectria flute canker in one forest in the foothills of the Canterbury plains.

Models developed to predict the potential range of Nectria flute canker in New Zealand suggest that the Canterbury plains are suitable for the disease, yet it appears that this location is far less suitable than Banks Peninsula. The present model is limited in that confirmed disease records in New Zealand cover a reasonably narrow climatic zone, particularly in terms of rainfall.

In order to better predict the effect of the pathogen when it reaches forests north of its present range, more data on the severity of the disease in *Pinus radiata* plantations growing in wet regions are needed. These data will become available through a new collaboration with Chilean researchers, which will enable New Zealand access to disease severity data from the wet regions of southern Chile.

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Pruning, branch diameter and stem disease

Nectria flute canker occurs almost exclusively in pruned radiata pine stands. The larger the branch at the time of pruning, the greater the probability of flute canker disease. Whilst flutes were common immediately after pruning, they tend to disappear with time, particularly where the pruned branch was less than 6 cm in diameter.

UNDERSTANDING NECTRIA

Scion undertook a project to further examine the relationship between branch diameter, pruning and the occurrence of disease by:

- examining records of disease recorded in radiata pine trials as part of a routine remeasurement,
- examining the predicted range in branch diameter at the time of pruning from TreeBLOSSIM simulations (a tree growth model that can predict branch development).

In the first part of the study, the diameter of the largest branch recorded in a pruning lift was compared with the incidence of disease (recorded as caused by either *Diplodia* or *Neonectria*) at a later age. The data came from radiata pine trials established to monitor the growth of radiata pine. Most were pruned in the mid to late 1990s and assessed in the mid to late 2000s.

There was no clear relationship between the diameter of the largest branch in a pruning lift and the incidence of disease at a later age. This result could have been anticipated because many of the trials were assessed before Nectria flute canker was known and there was no requirement to assess disease or damage to the stem. There could be much benefit from assessing current silvicultural trials for key diseases by well trained staff, even if the benefits of doing so are not readily apparent now.

Secondly, TreeBLOSSIM simulations predicted the range of pruned branch diameters for different initial stockings and different pruning regimes.

The TreeBLOSSIM simulations indicated that over a realistic range of initial stockings, few pruned branches would be larger than 6 cm.

This finding suggests that Nectria flute canker may not be a problem in many stands. However, further research to better understand branch development, and the relationship between branch diameter and the incidence of Nectria flute canker are recommended.

This is the first study in which TreeBLOSSIM has been used to examine branch diameters in pruning lifts.

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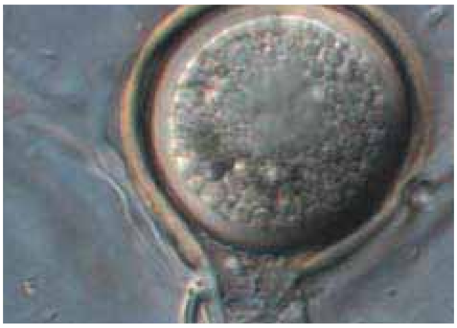
Does pruning induce Nectria flute canker?

Nectria flute canker is associated with *Neonectria fuckeliana*, yet the fungus has been found in unpruned trees with no disease. An experiment was established in December 2009 to further our understanding in the role of pruning stress and the initiation of disease development.

All stubs on three whorls on 180 trees were measured and assessed for fluting and *Neonectria* fruitbodies. Increment cores were taken to test for the presence of the fungus and these have now been processed. Trees are due to be reassessed for disease in March. Once results from the sampling and assessments have been compiled we will be able to test the hypothesis that pruning with the fungus present induces Nectria flute canker.

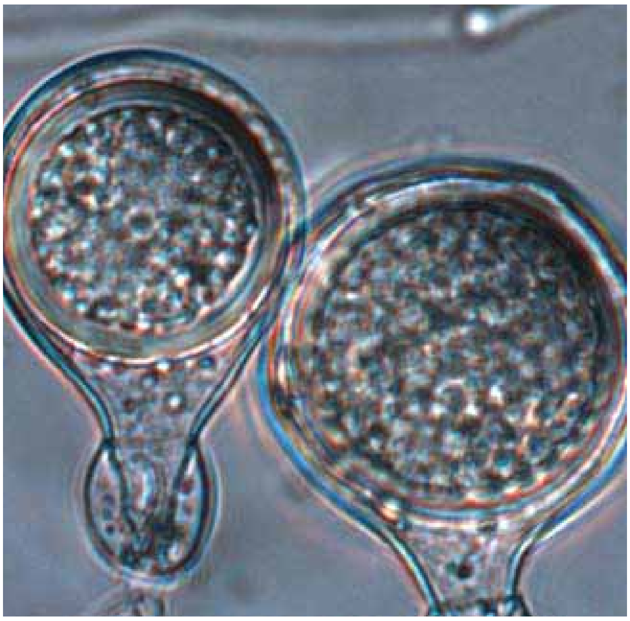
RAPID IDENTIFICATION OF PATHOGENS

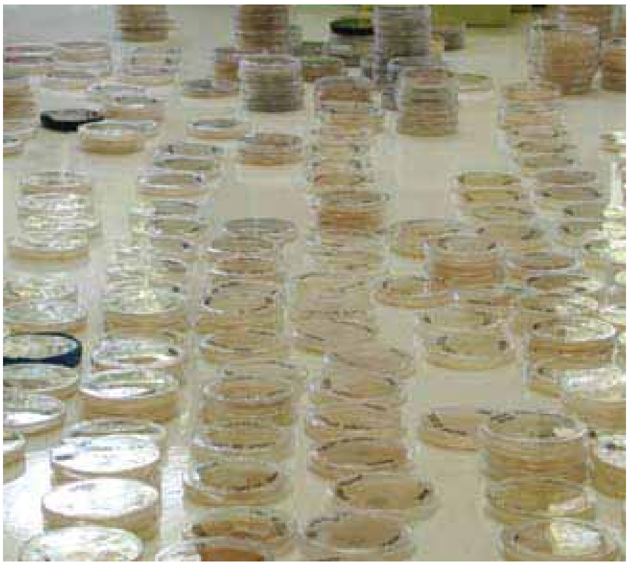
Macroarrays are a DNA based method that can allow several different pathogens to be identified in one simple screening procedure.



Microarrays can be used to rapidly detect a variety of organisms in samples ranging from environmental samples to diseased tissues. The key to the macroarray technique is the development of species specific "oligos" (i.e. short sequences of DNA) that are unique to the pathogens of interest. This step in the process can be time consuming and difficult, but macroarray printing and the experimental protocol are relatively easy.

Scion staff have developed species specific oligos to recognise eight different pathogens of radiata pine: *Dothistroma pini, Cyclaneusma minus, Neonectria fuckeliana, Diplodia pinea, Armillaria novae-zelandiae, Fusarium circinatum, Endocronartium harknessii (syn. Peridermium harknessii)* and *Phytophthora pinifolia*. An exciting result was that the method was able to detect





N. fuckeliana within radiata pine, demonstrating the ability of the test to work in infected plant material. Other pathogens are yet to be tested *in planta*.

To date, the experiments that have been conducted have focussed on proof of concept. Research is ongoing to quantify the specificity of the array, to ensure that it only detects the pathogens of interest, and the sensitivity of the array, to determine the detection thresholds. This research is being undertaken in collaboration with Dr André Lévesque, Agriculture and Agrifood Canada, who has developed macroarrays for plant pathogens.

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ECTOMYCORRHIZAL FUNGI

Ectomycorrhizal fungi are an intrinsic part of the rhizosphere and plant nutrition

Ectomycorrhizal (ECM) fungi enhance the growth of plantation species and facilitate the establishment of new tree stocks in the forest. ECM fungi have been a research focus at Scion in recent years, focusing on species identification with molecular methods and investigating the role of ECM fungi after trees leave the nursery and are planted out.

In the latest study we investigated ECM of seedlings of different stock (bare root, cuttings and root trainer stock) and hosts (*Pinus radiata* and *Pseudotsuga menziesii*) in five nurseries and during the first year of out planting in the five sites across New Zealand. The fungal species were identified with a combined approach of morphological and sequence analysis, an approach which had not been used on ECM fungi of *Ps. menziesii* previously.

Overall, relatively few ECM symbionts were found yet cuttings had higher species diversity. Most ECM fungi found to be important in the nursery remained in the plantation. Only a few non-nursery ECM were found one year after planting in the forest.

ECM colonisation and plant material varied between nurseries – plants with low ECM abundance and small root systems in the nursery appeared to stagnate later in the plantation.

The aim of this two year project was to create an inventory of 'what is there' for New Zealand's two most important plantation species. This knowledge is building the base for work on future sustainable forestry management practises by determining the influence of nursery fertiliser and fungicide applications on the growth of ECM fungi, a project that started in 2009 (funded by Future Forests Research).

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How long wood can be salvaged from storm-damaged trees

A project is underway to find out how long wood can be salvaged from storm-damaged trees before deterioration makes recovery uneconomic.

Current research is examining how both the season and the region influence the log recovery period.

Trees were felled early in 2010 to simulate storm damage at sites in Northland, the central North Island, Nelson and Canterbury. Further felling is being conducted in the same stands up to four times during the year to find out how the season in which a storm occurs influences the rate of wood deterioration. Felled trees are being monitored by periodically cutting discs along selected stems (destructive sampling) in order to determine how rapidly sapstain develops.

So far, the proportion of sapwood affected by stain has steadily increased in felled trees as the percentage moisture content has decreased. However, the rates of drying and deterioration appear to vary between regions. Values differ greatly between trees, but initial indications are that stems in Northland and Nelson may deteriorate more rapidly than those in the central North Island and Canterbury.

It also seems that predictions on the time available before sapstain develops in operationally harvested logs may not directly apply to storm-damaged stems which remain protected by a high proportion of intact bark. For instance, the national sapstain index predicts that sapstain will develop in about 40 days after harvest during early spring in Nelson but in an earlier study we found less than 10% of sapstain on average after more than 100 days in severed stems and after 1 year in uprooted trees. Four months after felling we observed little beetle attack (<5%) in both severed and still-rooted trees.

Further results are pending, and final outcomes will be reported under a commitment to provide a protocol to growers by June 2012. The overall project has the cooperation of a number of forest owners, and a significant part of the research involves an M.Sc. student at the University of Canterbury, who is also investigating the importance of bark beetles as potential vectors of sapstain fungi.

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An Australian parasitic wasp has been identified as a promising biological control agent for gumleaf skeletoniser.

The gumleaf skeletoniser is an Australian moth whose caterpillars have the potential to defoliate eucalypt plantations throughout New Zealand. Gumleaf skeletoniser is now widespread in the Auckland region, and is also present in Waikato, Coromandel and Bay of Plenty. The pest has recently been found in Hawkes Bay and Nelson and is likely to continue spreading.

Biological control of gumleaf skeletoniser

Scion received permission from the Environmental Risk Management Authority (ERMA) in 2010 to release a parasitic wasp into New Zealand aimed at controlling the gumleaf skeletoniser *(Uraba lugens)*. Biological control has been a safe and effective means of control for other eucalyptus pests in New Zealand.

The Australian wasp, *Cotesia urabae*, was found to be the most promising biological control agent because it will specifically target the gumleaf skeletoniser caterpillar. The wasp was obtained from Tasmania, with the help of Dr Geoff Allen (University of Tasmania/ Tasmanian Institute of Agricultural Research), so is adapted to a climate similar to New Zealand. A colony of wasps was raised in quarantine and the first official release was made in January 2011 in the Auckland Domain. The success of the release and ongoing effects will be closely monitored over the next few years.

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Re-establishing cleobora

The southern ladybird, *Cleobora mellyi* (Coccinellidae), is a predatory beetle that feeds on eucalypt and acacia pests. This beneficial insect was introduced to New Zealand from Australia in the 1970s and 1980s in the hope it would help control the eucalyptus tortoise beetle, *Paropsis charybdis*. Unfortunately it established only at one sheltered bay in the Marlborough Sounds at that time.

Cleobora mellyi also requires additional food sources to reproduce successfully, particularly in the form of sap-sucking psyllids, and these may not have been sufficiently abundant in the

1980s for the released ladybirds to be successful. Only two psyllid species were present on eucalypts in New Zealand when *C. mellyi* was introduced and at certain times of the year these species are scarce, potentially leaving ladybirds hungry. At least nine new psyllid species, as well as additional leaf beetle species have established in New Zealand on eucalypts and acacias since the original release of *C. mellyi*. Thus conditions were right to increase the distribution of this predator.

The discovery in 1996 of a new tortoise beetle, *Dicranosterna semipunctata*, on blackwood (*Acacia melanoxylon*) in New Zealand caused renewed interest in *C. mellyi* as it also feeds on this pest (now widespread in the upper North Island). In 2005 *C. mellyi* were collected from blackwoods and eucalypts in the Marlborough Sounds to try again to relocate this predator to other areas.

The collected beetles were reared in captivity then released at 19 sites in the North Island during 2006 and 2007 (Figure 1). This summer most of the sites have been checked for establishment of *C. mellyi*, with support from the Forest Biosecurity Research Council. *Cleobora mellyi* were found at nine of the 15 sites visited in the North Island, as well as at one of the two Southland release sites. At some sites establishment occurred from releases of only 23 ladybirds.

The southern ladybird is now established in the following regions, in addition to the Marlborough Sounds site: Northland (Kaitaia, Kerikeri), Auckland (Awhitu and Warkworth), Coromandel (Whangapoua), Waikato (Pirongia), Bay of Plenty (Rotorua), and Southland (Tokanui).

Although the search effort was concentrated on blackwoods, eucalypts were also examined at most release sites. Only one individual *C. mellyi* adult

Figure 1: Map showing the locations where *C. mellyi* is now known to be established, and sites where it has been released but not yet re-collected.

was found on eucalypts, suggesting they may have a preference for blackwoods and their pests. Further research is required to understand what effects *C. mellyi* can have on tortoise beetles and psyllids on eucalypts and blackwood.

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Improved *Trichoderma* formulations are being developed for forest nurseries to enhance the health and vigour of radiata pine cuttings and seedlings

Root promotion in cuttings

A high-throughput screen using *Impatiens* cuttings was developed to evaluate large numbers of *Trichoderma* isolates for their ability to enhance root initiation and growth. Over 100 *Trichoderma* isolates (from the TrichoBank™ collection) were screened.

The best performing *Trichoderma* isolates, including several novel ones, accelerated root initiation and growth and improved cutting survival and health. A new mixture of three *Trichoderma* isolates (WT3) and the natural product fulvic acid (FA) consistently performed well. WT3 and FA increased the survival of healthy rooted container grown (PF Olsen Nursery, Waiuku) cuttings by 12% and 13% respectively. In a soil bed nursery trial (Rangiora Nursery) WT3 and FA gave increases of 41% and 43%, compared with the untreated control. At the Te Ngae Nursery, FA substantially improved cutting survival (by 65%). Large scale (over 140,000 cuttings) forest nursery validation trials at the PF Olsen, Rangiora and Te Ngae Nurseries using the best *Trichoderma* treatments and FA were established in 2010.

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Induced resistance - *Trichoderma*

Plants possess a remarkable diversity of mechanisms to fend off attacks from pathogens, with preformed physical and chemical barriers forming the first line of defence. When a plant recognises a breach of the frontline it has the capacity to mobilise an impressive array of inducible defences to the infection point. All plants possess inducible defences, however, it is the speed and intensity of the induced response that determines whether or not the attempted infection is stopped. Disease occurs when this response is too weak and/or too slow.

Forestry Biocontrol Earns Bayer Innovator Award

Dr Robert Hill from the Bio-Protection Research Centre, Lincoln University, was awarded the 2010 Bayer Innovators Award for Agriculture and Environment, for his work with Trichoderma biocontrol.

Numerous studies have shown that topical application of certain beneficial micro-organisms and naturally occurring chemical 'elicitors' can be used to 'condition' plants so that their response to pathogen attack is enhanced. These plants are said to express 'induced resistance'.

Our studies on *Pinus radiata* have demonstrated that foliar spray with natural products and root application of *Trichoderma* spp., can result in stimulation of defence mechanisms in seedlings and enhanced resistance to diplodia dieback caused by *Diplodia pinea*. Furthermore, more recently our studies suggest that there may be some synergy or additive effect on pine defence when the treatments are combined. This approach offers some potentially exciting opportunities for the future.

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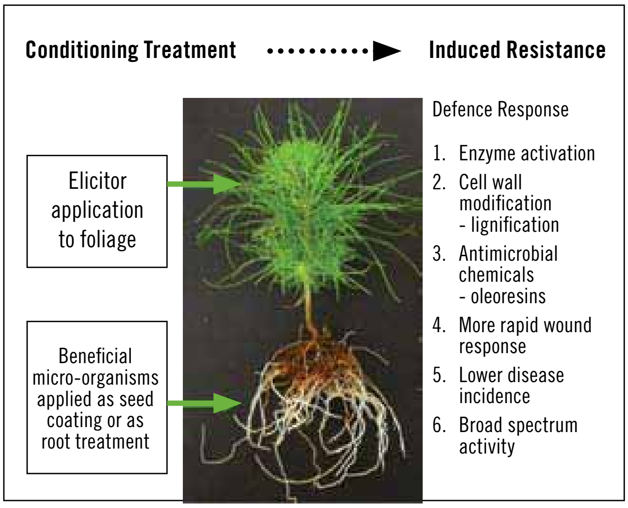

Beneficial entomopathogenic fungi

Progress has been made on optimum delivery systems for entomopathogenic fungi for bark beetle control. The programme has been investigating use of the fungi *Beauveria* and *Metarhizium*, both active against bark beetles in the laboratory. Preliminary research established that the fungi could be effective in the field, however formulation and application as biopesticides in plantation forestry situations was unlikely to be economically viable.

The discovery that *Beauveria bassiana* can become endophytic in pines changed the direction of the programme. Surveys of natural pine stands indicated infection rates in mature pines of over 20% and research has continued to demonstrate that the fungus can be introduced into seedlings through root dipping and seed coating and can persist in seedlings for several months.

The programme will now focus on the effect of endophytic fungi on insects feeding on trees. Research has also continued to determine the compatibility of entomopathogenic fungi with fungi used to control plant diseases, *Trichoderma* spp. It is an advantage to use fungi to control insect and plant diseases in seedlings, so the compatibility of the fungi has practical implications. Building on research that demonstrated no inhibitory effects in culture or for spore germination, large-scale pot trials were established in non-sterile soil. At high doses (above likely commercial applications) a decrease in *Beauveria* was found in the presence of *Trichoderma*, however this effect was not seen at lower, field-rate inoculum levels. These experiments are continuing in the presence of pine seedlings.

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Beneficial fungi beat fungicides in Borneo

A new bioprotection system using *Trichoderma* for *Acacia mangium* in the Planted Forest Zone of Sarawak was developed at the Samarakan Nursery for Grand Perfect and Sarawak Planted Forests. Pure *Trichoderma* cultures were isolated from a variety of healthy plant species in the Planted Forest Zone and evaluated in nursery trials with *A. mangium* seedlings.

The best performing *Trichoderma* isolates increased *A. mangium* seedling growth and vigour, and reduced disease losses without using any fungicide sprays. The proportion of seedlings "meeting specification for planting out into the forest" was increased by 66% using *Trichoderma* inoculation versus nursery standard practice (fungicides) for production of over 30 million *A. mangium* seedlings per annum.

Large scale validation using around 5 million seedlings, produced exactly the same results, (a 66% increase in trees "meeting specifications"). *Trichoderma* also reduced the length of the growing cycle by 20%. These results led to a decision late in 2009 to build a *Trichoderma* mass production facility on the nursery site and the removal of all fungicides from the seedling production programme.

The resulting increase in productivity from the nursery is estimated to be worth over NZ$3 million per year, a figure that does not take into account savings from fungicides or any environmental benefits. Pilot scale plantation trials using *Trichoderma* inoculated nursery stock have shown a significant reduction in mortality from diseases in the first year after planting in the forest.

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Forest Biosecurity Research Council

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For more information on FBRC, membership details and science reports.

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