An international project to study pitch canker in Radiata pine
Michael Devey\textsuperscript{1}, Colin Matheson\textsuperscript{1} and Michael Carson\textsuperscript{2}

Introduction
Pitch canker, caused by Fusarium circinatum (= Fusarium subglutinans f. sp. pini), is a disease of pines and other conifers. Apparently originating in Mexico (Wikler and Gordon 1999), it is endemic in forest nurseries and plantations in the SE USA (Blakeslee and Rockwood 1999), and has in recent years been reported in Japan, South Africa, and Spain (Muramoto et al. 1988; Viljoen et al. 1994; Dwinell 1999). In California, it first appeared in 1986 (McCain et al. 1987) and has rapidly spread through the three mainland populations of radiata pine (Pinus radiata). In forest nurseries in the SE USA and South Africa, the disease is known to cause "damping-off" and death of seedling pines. However, when provided with a suitable infection court, the pathogen can infect all parts of the tree, including needle and stem tissue, the main bole, roots, cones and seeds. The tree generally responds to infection by producing large amounts of resin and the formation of cankers in woody tissue, hence the common name (Figure 1). Already, in California, thousands of native radiata pine trees have either died from leader and bole damage caused by pitch canker, or have been removed due to their poor health, and the increased risk they represent for infection of neighboring stands (Mead 2000).

Although there is some evidence of direct infection of trees by the pathogen (Dwinell et al. 1985), pitch canker infection on either naturally-regenerated or planted pines is generally thought to result from insect wounds and vectoring of fungal spores (Storer et al. 1999). In California, there are numerous insect species known to be capable of transmitting the pathogen to radiata pine, such as species of Pityophthorus (twig beetles), Conophthus (cone beetles) and Ernobius punctatus (deadwatch beetle) (Adams 1997; Dick 1998; Hoover et al. 1995, 1996; Gordon et al. 1997). Species of Rhacionia (shoot tip moth) cause damage to loblolly pine in SE USA, causing wounds which are readily infected with pitch canker (Blakeslee and Rockwood 1999). The deodor weevil, Pissodes nemorensis, has frequently been found carrying the fungus in Florida and infection of pine seedlings has been demonstrated in laboratory tests with artificially contaminated weevils (Blakeslee et al. 1978). Chilean plantations of radiata pine suffer serious damage from the pine shoot tip moth, Rhacionia buoliania, and there is understandable concern that the introduction of pitch canker could make things much worse. Neither New Zealand nor Australia currently has Rhacionia species present, but past depredations caused by Sirex noctilio in both countries, and the presence of other serious timber boring insects (e.g. Ips spp. in Australia), indicate that some contingency investment in pitch canker research is advisable.

Radiata pine is one of the most important timber species worldwide, with more than 4 million hectares of plantations, principally in Australia (0.8 million ha), New Zealand (1.5 million ha) and Chile (1.4 million ha). Among the pines, radiata is one of the most susceptible species to pitch canker (Hodge 1999). If the disease were to be introduced, it could present a significant economic threat to the forest industry in these countries.

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\begin{figure}[h]
\centering
\includegraphics[width=0.8\textwidth]{resin_flow_pitch_canker.png}
\caption{Resin flow from pitch canker infected radiata pine in Monterey, California.}
\end{figure}
The Research Venture

In response to forest industry concerns, a broad-scale research project called 'IMPACT' was initiated to study pitch canker. The project is being managed by CSIRO Forestry and Forest Products. The Southern Tree Breeding Association in Australia, the New Zealand Radiata Pine Breeding Company (NZRPBC) and Forest Research in New Zealand, and Controladoras de Plagas Forestales S.A. in Chile represent the radiata pine industry in the three radiata-growing countries. Each organisation represents from 10 to 20 companies. Other collaborators in California include the USDA Forest Service, Institute of Forest Genetics in Placerville; Tom Gordon at the University of California, Davis; and the Pebble Beach Company in Monterey. The IMPACT project has benefited greatly from the assistance of the Pitch Canker Task Force, currently headed by Steve Staub, which was set up to develop management guidelines and define research priorities for pitch canker in the Monterey pine forests (http://frap.cdf.ca.gov/pitch_canker).

The IMPACT Monterey Workshop was organised to initiate the project in December 1998. This was the first international gathering specifically designed to address the subject of pitch canker in Pinus radiata. Appropriately, the workshop was held at the Asilomar Conference Center in Monterey, among radiata pine trees under severe attack by pitch canker. The workshop included a number of research presentations on the disease followed by discussions to work out details for management and organisation of the IMPACT project. Proceedings of the workshop are available from CSIRO.

Strategy

The principal objectives for the project are to determine the level of resistance to pitch canker among elite lines and native populations of P. radiata, to estimate heritabilities and other breeding parameters for resistance, and to screen and develop disease resistant/tolerant planting stock for the future. Seed from

<table>
<thead>
<tr>
<th>Country/Organisation</th>
<th>Number of seedlots</th>
<th>No. of seedlings</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Half-sib</td>
<td>Full-sib</td>
</tr>
<tr>
<td>Australia</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Southern Tree Breeding Association</td>
<td>-</td>
<td>100</td>
</tr>
<tr>
<td>CSIRO</td>
<td>-</td>
<td>63</td>
</tr>
<tr>
<td>New Zealand</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Radiata Pine Breeding Company</td>
<td>141</td>
<td>-</td>
</tr>
<tr>
<td>Chile</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Controladora de Plagas Forestales</td>
<td>160</td>
<td>-</td>
</tr>
<tr>
<td>Controls</td>
<td>4</td>
<td>-</td>
</tr>
<tr>
<td>Total</td>
<td>305</td>
<td>163</td>
</tr>
</tbody>
</table>
Australia, New Zealand, and Chile was sent to the Institute of Forest Genetics (IFG) in Placerville where it has been sown for subsequent evaluations in glasshouse and field trials.

**Genetic Material**

A total of 531 seedlots have been received at IFG; Det Vogler and Annie Mix are overseeing the project there (Figure 2). Samples of all of the seedlots have been sown once, and in some cases twice, where there were fewer than 20 germinating seedlings. As of August 2000, there were 440 seedlots with between 20 and 30 seedlings each.

The material includes a large number of half-sib (OP) and full-sib crosses as well as collections from the native populations (Table 1). The material from Australia includes 163 full-sib crosses of elite lines; STBA and CSIRO coordinated their contributions so that they included a structured diallel mating design to estimate breeding parameters. There are also 63 seedlots from the five native California populations collected by Ken Eldridge and Tony Firth back in 1978, and which had been in storage in Australia. Three of these samples were bulks from the Cambria population, and the remainder were individual tree collections from Ano Nuevo, Monterey, Cedros and Guadalupe. The New Zealand and Chilean representatives each contributed 141 and 160 OP seedlots, respectively. The NZRBC seedlots represent half-sib OP progenies of the best breeding parents currently represented in clonal seed orchards. These OP seedlots will be screened to provide parental breeding values for pitch canker resistance, from which full-sib crosses with both parents resistant may be developed. The NZRPBC also contributed seed to be used for controls. The controls are needed to standardize inoculation results over time, since it is known that the response to the pathogen varies with temperature, and inoculations will be done over a period of several months.

**Glasshouse Inoculation**

It was decided at the IM-PACT Workshop that a glasshouse evaluation for resistance to pitch canker should precede any large-scale field testing. There are a number of reasons for this:

Because the overall level of resistance to pitch canker is expected to be quite low, field trials to test a wide range of material could be mostly dead within 12 months of planting.

A field trial in California would expose the pine seedlings to only a few of the known strains (or "vegetative compatibility groups") of the causal fungus, yet other, potentially more virulent strains of *Fusarium* could be introduced to Chile, New Zealand, or Australia. Glasshouse screening allows for a range of pathogen strains to be used (although in the initial screening, only a single local fungal isolate will be used).

The insect vectors for the disease differ in different countries in which pitch canker has been identified, so there is a need to provide a screening result for fungal resistance that is independ-

![Figure 3: Distribution of pitch canker in California, also showing locations of three mainland populations of Pinus radiata (Ano Nuevo, Monterey and Cambria) and Placerville where the seedlings are being grown. Pitch canker data from Adams et al. 1999.](image-url)
ent of the relevant insect vector.

Glasshouse screening methods for assessing pitch canker resistance have been developed both by Tom Gordon at UC Davis and at the USDA Resistance Screening Center in Asheville, North Carolina. These procedures may prove to be more efficient than field tests in ranking seedlots for their resistance.

Glasshouse screening is expected to give earlier results than field screening.

To avoid a risk of introducing the disease outside of the present zone of pitch canker infestation, inoculations for the glasshouse trial will not be done in Placerville. The Pebble Beach Company in Monterey has offered space in their glasshouse for seedlings to be held during the artificial inoculation and early development stages. At about age 12 months, the seedlings will be transferred the 300 km distance from Placerville to Monterey (Figure 3). Once in Monterey, the seedlings will be retained at this location, and destroyed following further evaluations for the disease.

Tom Gordon and Det Vogler will coordinate the inoculations. Depending on the capacity of the glasshouse, the seedlings will be inoculated in lots of about 2500. Each lot of 2500 seedlings will be inoculated during a two-week interval and scored 5-6 weeks later. A local F. circinatum isolate endemic to the Monterey Peninsula will be used for the inoculations. A small hole (1.6 mm) will be drilled in one of the branches and 5 microlitres of an aqueous spore suspension will be placed in the hole. Presence or absence of lesions caused by pitch canker will be noted after 5-6 weeks, and seedlings without lesions will be inoculated a second time. The data obtained will be lesion length for each tree, as previous experiments have shown that this is a good predictor of susceptibility based on subsequent field evaluations (Gordon et al. 1998). Seedlots will then be evaluated and ranked for their relative resistance to pitch canker, as predicted by lesion length in the glasshouse study.

Field Trials

Based on results from the glasshouse evaluation, a subset of the radiata pine seedlots will be selected for planting in field trials at two locations within the zone of pitch canker infestation in California. The seed will again be sown at IFG beginning about January 2002 for planting 12 months later. Locations for the field trials have yet to be identified. To avoid possible contamination with pollen of unknown origin, the trial sites will need to be some distance away from the native populations of P. radiata. A site was proposed at the Swanton Pacific Ranch near the southern end of the Ano Nuevo population, which is exposed to prevailing winds from the northwest. However, upon subsequent consideration, it was decided that this site might be too close to native P. radiata stands in the vicinity. In preliminary discussions with Robert Reid, the City Forester in Monterey, it has been suggested that one or two sites may be located in Monterey. These would be well away from existing native stands. It would be desirable if the second field trial site were located in some other area, perhaps near Cambria, for evaluations of different environments and races of the pathogen. The project team will be evaluating this possibility.

The IMPACT glasshouse and field studies should provide indicative rankings of the relevant seedlots for their natural genetic resistance to the pitch canker disease. Using these results, it will be possible for breeding programmes in the participating countries to identify and develop breeding crosses with increased resistance, for use in the event of an accidental introduction of the disease. Similar screening procedures may also be used to identify and rank individual clones of radiata pine with resistance, also for direct use in future plantations. In the (expected) event that each participating country will identify breeding parents with useful resistance, there will also be an opportunity for them to exchange breeding material (pollen, seeds, or vegetative material) in order to enhance resistance in participating company plantations.

Future Studies

The IMPACT field trials will provide a resource for research on pitch canker over the next 10 to 15 years. Research projects could be undertaken independently or in collaboration with supporting organisations. Some of these studies may involve pathogenicity of the fungus, mode of transmission and risk analysis, estimation of heritabilities and other genetic parameters, and the use of DNA marker methods for genetic mapping of quantitative trait loci controlling resistance, leading to marker-aided-selection of pitch canker resistant clones or seedlots of radiata pine. As a result of this project, it is hoped that the cooperating countries will continue to work together to develop common biosecurity strategies and standards for effective protection of pine plantations against other forest pests and diseases.

Acknowledgements

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