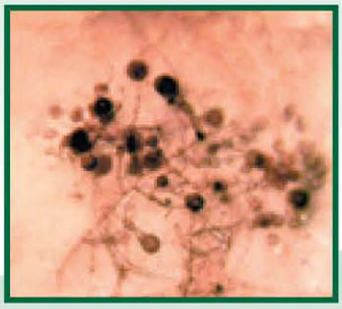

Rainforest Dieback Mapping and Assessment

Edited by P.A. Gadek
and S. Worboys

Rainforest CRC
Cooperative Research Centre for Tropical Rainforest Ecology and Management

RAINFOREST DIEBACK MAPPING AND ASSESSMENT

Phytophthora species diversity and impacts of dieback on rainforest canopies

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Rainforest CRC

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June 2003

TERMS OF REFERENCE

This publication provides a report on the status of dieback studies in upland rainforest of the Wet Tropics World Heritage Area of North Queensland. The report focuses on field work undertaken over the period August 2001 – July 2002, and represents Stage 3 in a program of studies initiated in 1998. Funding has been provided by the Wet Tropics Management Authority, initially through the Rainforest Cooperative Research Centre. Studies over the past four years have been designed to address the following Terms of Reference.

STAGE 1 – 1997

A preliminary review (Gadek 1997), prepared in response to concerns from the tourism industry, identified the need for further studies in to the status and distribution of *Phytophthora cinnamomi* in the upland rainforests of the Wet Tropics. A key recommendation in this review was for a workshop of invited experts to discuss the issue. This workshop was held in April 1998 and the resulting proceedings has been published (Gadek 1999).

STAGE 2 – 1998-2000

These terms of reference were set down for works undertaken in the period 1998-2000. Outcomes from these studies were reported in Gadek *et al.* (2001).

Purpose of Contract

To determine the current distribution and extent of health of rainforests in areas identified as suffering from forest dieback using a combination of physical surveys, remote sensing and GIS techniques.

Objectives

- To systematically examine and markup rainforest dieback air photo patterns contained within parts of the area identified as displaying symptoms of canopy dieback.
- To undertake sufficient field surveys to characterise and ecologically describe the observed photo patterns including an assessment of the ecological health, successional status and preliminary identification of susceptible and resistant species.
- To undertake an assessment of the feasibility of remote sensing/GIS technology for determining the spectral characteristics of known instances of patch death and their applicability for detailed forest health and ongoing monitoring of the phenomenon.

Tasks to be Performed

a) Detailed aerial photographic markup and interpretation of observable canopy dieback including accurately locating and marking sufficient control points necessary for accurate digital pattern transfer and rectification.
b) Undertake fieldwork in a stratified manner which will allow description of both within and between polygon variation. Descriptions will include the structural and physiognomic characteristics of representative photopatterns and provide site descriptions and floristic lists (at least of canopy species) of representative locations. Basic site descriptions and species inventories will follow a standard and consistent format.
c) Undertake an assessment of the utility of remote sensing technology in conjunction with GIS methodology for determining the spectral characteristics of forest dieback patches identified in a) and b) above, and their applicability for detailed forest health assessment and on-going monitoring of the phenomenon.

STAGE 3 – 2001-2002

Specific recommendations arising from the 1998-2000 studies (Gadek *et al.* 2001) are listed below.

- Continue to systematically examine and markaup rainforest dieback patches from air photopatterns contained within parts of the area identified as displaying symptoms of canopy dieback.
- Continue to undertake field surveys to better characterise and ecologically describe the observed photopatterns including an assessment of the ecological health, successional status and identification of susceptible and resistant species; and
- Apply remote-sensing/GIS technology to a detailed assessment of forest health and on-going monitoring of canopy dieback in the Wet Tropics World Heritage rainforests.

Specific tasks:

a) Develop specific spectral signatures for dieback patches from airborne video (Louis *et al.* 1995) and LandSat ETM+ data and apply them to the whole study area.
b) Assess canopy variance using spectral signatures from a random sample of equivalent areas of health forest as a basis for the development of protocols for routine monitoring using remotely sensed data.
c) Establish protocols to determine dieback susceptible and non-susceptible areas of the Wet Tropics World Heritage Area.
d) Determine whether current findings apply across other affected communities in the Wet Tropics, and particularly if the association with drainage lines and roads highlighted in the Tully Falls Region is repeated in other regions of the Wet Tropics.
e) Undertake aerial surveys and monitor established field plots to track the occurrence and extent of canopy dieback over time.
f) Determine the distribution of *P. cinnamomi* in the Wet Tropics, using the sampling strategies developed here, and determine if *P. cinnamomi* is limited to a particular environmental stratum.
g) Establish causation (rather than inference) between the presence of *P. cinnamomi* and tree deaths in rainforest communities, and determine the triggers to virulence in rainforest ecosystems.
h) Provide an assessment of genetic diversity within and between populations of *P. cinnamomi*, particularly within tropical rainforest ecosystems.

STRUCTURE OF THE REPORT

The following report presents outcomes of research undertaken in the period 2001-2002.
Section 1 reports the outcomes of investigations into the distribution and identification of *Phytophthora* species in the Wet Tropics, and the genetic structure of populations of *Phytophthora cinnamomi* at selected sites. This section addresses specific task (h) in the terms of reference.

Section 2 discusses research into the relationships between the physical and biological environment and patch death in the forest, and is to be read in association with *Rainforest Dieback: Assessment of the Risks Associated with Road and Walking Track Access in the World Heritage Area*, by Worboys and Gadek (2002). This section reports outcomes of studies designed to address recommendation (2) and specific tasks (d), part of (e), (f), and (g).

Tasks (a) and (b) are subject of a third report to be prepared by Professor David Gillieson and others. Protocols for determining dieback-susceptible and non-susceptible areas in the World Heritage Area are discussed in Worboys and Gadek (2002).

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ACKNOWLEDGEMENTS

This project would not have been possible without the willing assistance of many people who have shown a keen interest in this research, and invariably expressed concern as to the possible impacts of *Phytophthora* related dieback.

Planning for this project was facilitated by discussions with Will Edwards, Steve Goosem, Jill Landsberg, Steve McKenna, Garry Werren and Peter Stanton. Peter Stanton and David Stanton assisted with locating study sites.

Mike Stott and Terry Webb of the Wet Tropics Management Authority generously gave their time, and loaned aerial photographs for use in this project.

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Nigel Tucker and volunteers at the Centre for Tropical Restoration provided significant assistance and advice in the propagation of native rainforest tree cuttings. CTR was also extremely generous in its provision of space for cutting propagation. CTR and Bill Sokolich supplied seedlings for experimental use. Claudia Morris and Kerry Walsh assisted in obtaining permits for carrying out this work.

Lynton Vawdrey, Senior Experimentalist at the Department of Primary Industries, South Johnstone, offered to provide space in their Quarantine glasshouse for infecting plants with *Phytophthora* isolates.

Numerous field assistants gave their time, usually quite freely, including Bruce Wannan, Julia Goulding, Jair Schalkwijk and Michael, James Hill, Earl Davis, Paul Drake and Alex Cobb. Rob Jago provided expert advice on plant identification in the field.

Gavin Kingston and Rupert Russell guided us to isolated dieback sites on the southern end of the Carbine Tableland.

David Gillieson provided assistance and advice in the use of GIS applications, and created GIS layers of high and moderate susceptibility zones, and of *Phytophthora* sampling points.

Funding for the research was provided by the Wet Tropics Management Authority

EXECUTIVE SUMMARY

RESEARCH OBJECTIVES

- To develop and test a rapid and reliable identification protocol to ensure the accurate identification of *Phytophthora* isolates in rainforest soils;
- To describe the distribution of *Phytophthora* species within tropical rainforest ecosystems of Far North Queensland;
- To provide an assessment of genetic diversity within and between populations of *P. cinnamomi* within tropical rainforest ecosystems of Far North Queensland.

KEY FINDINGS

- Morphological techniques alone are inadequate for the accurate identification and determination of the distribution of *Phytophthora* species in the Wet Tropics Region. Due to these limitations, molecular protocols were developed that allowed an accurate and relatively rapid assessment of *Phytophthora* species diversity in rainforest soils. At least 5 species of *Phytophthora* are confirmed as present in soils within the Tully Falls / Koombooloomba Dam and Mt Lewis regions, and a more accurate description of the distribution of these species in these regions has been determined.

- *P. cinnamomi* was the most common species of *Phytophthora* in all regions surveyed. *P. cinnamomi*-related canopy dieback is widespread in the Wet Tropics. However, the presence of this pathogen is not always related to canopy dieback, as first noted by Brown in Gadek (1999).

- The level of innoculum of *P. cinnamomi* was greater at the scale of canopy dieback (more isolates were detected in affected than unaffected sites) as well as at the regional scale (more isolates were detected at Mt Lewis than Tully Falls / Koombooloomba Dam region).

- A preliminary determination of the genetic structure of the Mt Lewis populations of *P.cinnamomi* did not explain the appearance of virulent outbreaks of secondary symptoms of the disease– canopy dieback patches – in this region. Genotypes of *P. cinnamomi* in affected sites are significantly different to those that occur in unaffected sites, but further analysis of the data suggested that populations may be geographically structured. Further research is required to determine if virulent strains of *P. cinnamomi* exist and are correlated with canopy dieback, or if an antagonistic reaction between genotypes explains these virulent outbreaks.

- The observations of Gadek *et al.* (2001) on relationships between the distribution of mapped dieback polygons and roads, granite or rhyolite soils and notophyll forest type (although with a small number of incidences outside these areas) have been reinforced in this study for both mapped dieback polygons and unmapped dieback patches.

- The impact of *P. cinnamomi*-related dieback on some sites is dramatic and severe, in others the impacts are more diffuse and difficult to define. Although canopy openness is significantly affected by dieback, there was no consistent differences of canopy species diversity, living stem density or living stem basal area between affected and unaffected sites. However, dieback –affected stands have a higher density of field-susceptible species, which may contribute to the intensity of the disease on these sites.

- In severely affected sites, it appears most species in the community are affected, however, where the outbreak is less severe, we have recorded several species which appear more susceptible to attack, and others which display a form of field resistance. Preliminary lists of field susceptible and field resistant species, based on analyses of the complete dataset from this study, are provided in this document, and supersede any lists previously distributed. It is hoped these lists will be utilised in selecting species for planting in high-susceptibility zones.

- The absence of long-term monitoring data prevents us from drawing conclusions about the implications of *P. cinnamomi*-related dieback in the forests of the Wet Tropics World Heritage Area. We do not know, for instance, the characteristics of forest recovering on old patch death sites, or if the drought of 2002 affected the apparent of severity of dieback. Although no species or communities were found to be threatened by *P. cinnamomi*-related dieback, the long term threats cannot be determined from the existing information. A precautionary approach to management of *P. cinnamomi* is therefore required.

RECOMMENDATIONS FOR MANAGEMENT

Management recommendations for works in high-susceptibility zones of the Wet Tropics are given in Worboys and Gadek (2002).

A Technical Workshop organized by WTMA in March 2003, based on Worboys and Gadek (2002) and this report, identified a number of management issues including:

- Prevention of soil transport, particularly from lowland forests to high mountain tops;
- Introduction of wash-down procedures;
- Use of local seeds or plants from accredited nurseries;
- Use of certified *Phytophthora* free gravels in maintenance activities;
- The reinstatement or establishment of effective drainage on roads and walking tracks to prevent pooling of water.

FURTHER RESEARCH

Future research should focus on further understanding the distribution of *Phytophthora* species; and investigate the role of other *Phytophthora* species in natural tropical ecosystems.

In relation to *P. cinnamomi*, further research should:

- Continue canopy dieback mapping and monitoring;
- Establish if *P. cinnamomi* is native or an exotic introduction;
- Continue to investigate triggers which cause the manifestation of canopy dieback.

Dieback on high-altitude ridgelines

A question which has arisen during the course of field work in this project is the perceived relationship between patch-death and high-altitude ridgelines (Section 2.3.3). Dieback is widespread along the western access track to the summit of Mt Bartle Frere, on the Main Range north of Mossman, near the summit of Black Mountain (Harris Peak) north of Kuranda and in the rarely visited Mt Mackay area northeast of Mt Molloy. All of these areas are in REs 7.12.19, 7.12.20 or 7.12.16. It is not known if this correlation is real, or simply an artefact of biased observations (most highland walking tracks follow ridgelines, leaving hillslopes and valleys unsampled), nor have any *Phytophthora* species been isolated from soils except for Mt Mackay. There may be some environmental factors, such as poorly drained soils or susceptible species, present along ridgelines which predispose these environments to dieback.

The observation is of significant concern. These REs are home to a high number of endemic rare and threatened species which occur in these two REs (Sattler and Williams 1999). These ridgelines are utilised by recreational walkers and land managers (and wildlife?), all of whom have the potential to spread *Phytophthora*. These areas must be considered an extremely high priority for:

- confirmation of the presence of the pathogen by soil isolations,
- establishment of monitoring programs, and
- implementation of impact mitigation measures such as user education, restriction of access to areas away from the main route and/or closure of less used tracks (in consultation with user groups).

SECTION 1 – IDENTIFICATION AND DISTRIBUTION OF *PHYTOPHTHORA* SPECIES AND GENETIC POPULATION STRUCTURE OF *PHYTOPHTHORA CINNAMOMI*

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1.1 INTRODUCTION

While there is conclusive evidence that *P. cinnamomi* is the cause of canopy dieback in other Australian ecosystems (Weste 1974), similar evidence does not exist for the canopy dieback patches found in Far North Queensland. Nevertheless, the correlation between the detection of *P. cinnamomi* in soils and canopy dieback are clear and unambiguous. The pathogen can be isolated from dieback-affected forest as well as from seemingly unaffected sites (Brown in Gadek, 1999) where the expression of infection may be cryptic.

THE DISEASE TRIANGLE

In any disease system, three factors interact to govern the severity of the disease: the presence of a virulent pathogen, the presence of a suitable environment, and the presence of a susceptible host (Erwin & Ribeiro 1996) (Figure 1.1).

As these factors interact the severity of disease may increase or decrease accordingly (Erwin & Ribeiro 1996). In previous studies the approach has been to examine the host and the environment (Brown in Gadek, 1999; Pryce 2000), which revealed a correlation between disease severity and environmental factors. Variation in disease severity may also be the result of variation in virulence between strains of *P. cinnamomi*, and an investigation of this aspect is the subject of this report.

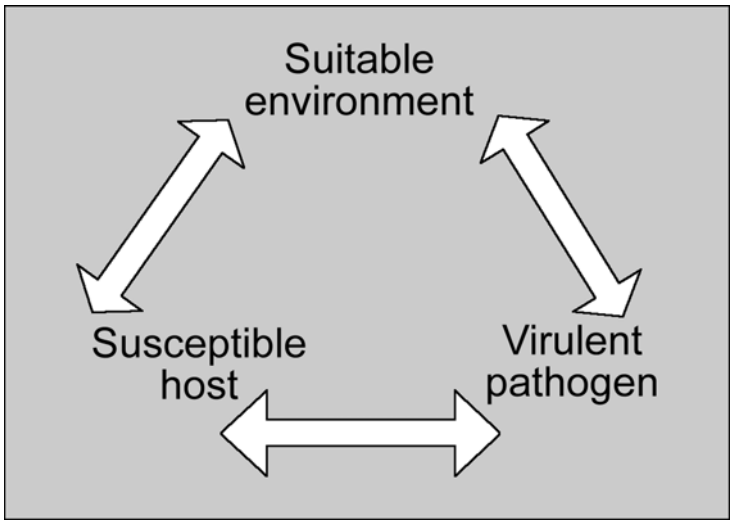

Figure 1.1. The disease triangle – three factors which interact to govern the severity of disease.

THE PATHOGEN - *Phytophthora cinnamomi*

Phytophthora (Family Pythiaceae, Order Peronosporales, Class Oomycetes, Kingdom Chromista), is a genus of fungus-like plant pathogens (Brasier 1992). The name *Phytophthora* means plant destroyer, and there are over fifty different species that will cause plant disease (Erwin & Ribeiro 1996). The most notorious of these is *Phytophthora cinnamomi*, currently distributed in over sixty-five countries worldwide including Australia.

THE PROCESS OF PLANT INFECTION

P. cinnamomi has a broad range of hosts and the ability to cause disease in over one thousand different species of plants (Erwin & Ribeiro 1996). In all of the species examined in detail, the primary symptoms of infection are the same (Cahill in Gadek,1999). Infection tends to occur at the unsuberised zone of elongation of the root tips (although it is also possible where tap and lateral roots join), at subterranean stomata or at fresh wounds in the collar of the trunk (Cahill in Gadek, 1999). As the mycelium grows through the stele, the cambium and phloem are destroyed while the metaxylem is generally left intact (Weste & Marks 1987). The associated tissue necrosis produces a characteristic red-brown discolouration or lesion (Weste & Marks 1987), which first appear as water-soaked areas.

These are the **primary symptoms** of infection and can occur in the roots or collar of the trunk. Irwin *et al.* (1995) reported that this root and/or collar rot results in

- leakage of electrolytes (due to increased permeability of the root membrane),
- an increase in the respiration rate,
- a reduction in the net hydraulic conductivity with related decreases in
    - mineral concentration within plant tissue;
    - root relative water content;
    - leaf water potential; and
    - transpiration.

The damage to the root system causes **secondary symptoms** of infection, which can include chlorosis (yellowing), wilting, and ultimately death of foliage and shoots (Irwin *et al.* 1995), and resultant canopy dieback. Infection by *P. cinnamomi* may not be visible if only primary symptoms occur.

PATHOGENICITY AND VIRULENCE OF *P. CINNAMOMI*

Pathogenicity is defined here as the ability of a pathogen to cause disease, while virulence is defined as the severity of disease that is caused by a pathogen.

In all plant species so far examined, zoospores of *P. cinnamomi* are attracted to the roots, and are able to encyst and penetrate the root tissue (Weste & Marks 1987). Although *P. cinnamomi* appears to be universally pathogenic, the severity of disease it causes (virulence) varies. Anatomical barriers to infection may be produced in species that are said to be resistant. These can include physical barriers such as callose deposits (Weste & Marks 1987) which may act to seal lesions off (Cahill & Weste 1983), lignin (Cahill *et al.* 1989), and/or the formation of a necrophylactic periderm, which restrict the growth of the pathogen (Tippett & Hill 1984). Despite these barriers, the pathogen generally remains viable and is able to reproduce (Cahill *et al.* 1989).

There may be a difference between susceptible and resistant species at the cellular level (*e.g.* Jang & Tainter 1990, Cahill & McCoomb 1992). A continuous range of resistance and susceptibility levels was found for some species of Jarrah (*Eucalyptus marginata*) (Stukely & Crane 1994). Thus, some hosts of *P. cinnamomi* are said to be resistant even if infection can occur.

GENETIC VARIATION OF *P. CINNAMOMI*

Isozymes were the first biochemical technique used to study the genetic variation of *P. cinnamomi*. This method detects some of the variation in DNA sequences which code for the specific proteins assayed. A number of isolates from a range of hosts and locations were collected in Australia and Papua New Guinea (Old *et al.* 1984). Using isozymes from 183 isolates, the difference between the two compatibility types (A1 and A2) were detected. In the Australian isolates there was low genetic variation and only three multilocus isozyme

types were found, two of A2 type and one of the A1 compatibility type. However, more genetic variation was found for the A1 compatibility type from Papua New Guinea in comparison to the isolates in Australia. A similar study was later performed in Australia in locations where both of the mating types could be found together in the soil (Old *et al.* 1988). No sexually-recombinant isozyme types were found in that study.

Only three multilocus genotypes were found when microsatellite molecular techniques were used on isolates collected from a number of locations within Australia (Dobrowolski 1999). These genotypes corresponded with the three isozyme genotypes found by Old *et al*. (1984). The three genotypes are thought to be distinct clonal lineages of *P. cinnamomi*. To date, the genetic variation that has been detected for *P. cinnamomi* in Australia is consistent with the presence of three apparently clonally reproducing lineages (Dobrowolski 1999). Unfortunately this study **did not include isolates from localities in Far North Queensland.**

GENETIC BASIS OF VIRULENCE

In a number of pathogens virulence has been shown to have a genetic basis (*e.g.* Hermans *et al.* 2000, and Baele *et al*. 2000 discuss this phenomenon in bacteria; de Wet *et al.* 2000; Smith & Stanosz 1995, and Kolmer & Liu 2000 represent recent studies of the phenomenon in true fungi). In order to correlate the occurrence of particular genotypes of *P. cinnamomi* with affected sites and to test whether those genotypes may be responsible for the virulent outbreaks of disease within those sites, an accurate identification and distribution of *P. cinnamomi* is required.

OBJECTIVES OF THIS STUDY

This section is divided into three parts, addressing the following objectives:

- To produce a rapid and reliable identification protocol to ensure that identification of the *Phytophthora* isolates is accurate;
- To determine the distribution of *Phytophthora* species within rainforest ecosystems in two regions of Far North Queensland, Mount Lewis and Tully Falls/Koombooloomba Dam, and to compare the results to those by Brown (in Gadek 1999) and (Pryce, 2000);
- To determine if the occurrence of canopy dieback patches in the tropical rainforests of Far North Queensland can be explained by the presence of particular genotypes, possibly highly virulent strains, of *P. cinnamomi*.

1.2 MORPHOLOGICAL AND MOLECULAR IDENTIFICATION OF *PHYTOPHTHORA* ISOLATES

1.2.1 INTRODUCTION

All previous studies of *Phytophthora* from Far North Queensland have used traditional morphological identification techniques. The traditional method of identification requires reproductive structures (*e*.g. oogonia, sporangia) to be available. Waiting for these structures to be produced in cultures can be extremely time-consuming, especially when dealing with large sample sizes. Even if produced, these structures can still be difficult or impossible to interpret. In an attempt to overcome problems of accuracy of *Phytophthora* species identification, molecular techniques are becoming more widely used. Where accurate identification is required, molecular results are less subjective and do not rely on particular characters that only occur in one stage of the life cycle. They are accurate because they have been developed to be species-specific, and are based on regions of the genome that show no within-species variation.

The following section reports the development of a rapid, reliable and cost-effective identification protocol by combining molecular and morphological identification techniques. This ensures that interpretations of distribution and genetic variation of *Phytophthora* species isolated in this study are more accurate and informative than previous studies have achieved.

1.2.2 METHODS

Soil sampling

Two areas were sampled: Mount Lewis and Tully Falls/Koombooloomba Dam. Sixteen (16) soil samples, each with approximately 250 g of soil were collected from each of twelve sites at Mount Lewis, and eight samples from each of twelve sites at Tully Falls/Koombooloomba Dam. These were stored in plastic clip-seal bags and returned to the laboratory for processing.

Lupin baiting technique

Lupinus angustifolius L., Fabaceae, (blue lupin) is a plant species particularly susceptible to infection by *P. cinnamomi* (Chee and Newhook 1965). A technique has been developed for baiting *P. cinnamomi* out of the soil using lupins (Pratt and Heather 1972) and may also be useful for the baiting of some other *Phytophthora* species (Drenth and Sendall 2001).

The roots of newly germinated lupins (seeds sourced from Mr Donald Spurs of Rockfield Pty Ltd, Latrobe, Tasmania) were placed in a 1:10 soil/water mix, which allows the zoospores to locate and infect the roots. After symptoms of infection were detected in lupin seedlings (i.e. lesions on the roots, wilting of the leaves), root sections were taken. The roots were removed from the water and surface sterilized by immersing for 2 minutes in 70% alcohol (Drenth and Sendall 2001). They were then dried with sterilised paper towelling and cut into 0.5 cm sections. The root sections were placed onto an agar growth medium designed to be selective for *Phytophthora*, and containing antibiotics. This medium discourages growth of fungi and bacteria, so that a pure culture of the target organism can be obtained. After two days, the plated root sections were examined for growth of *Phytophthora*. When mycelium was detected, subcultures were taken and plated onto selective medium. Three subculturing rounds were performed.

Since it is possible that one root section culture may comprise more than one individual or species (there may have been several infections of the same root), a hyphal tip subculturing technique was used. The tip of an individual hyphal strand was located with the aid of a stereomicroscope, removed and placed onto a growth medium (10% Campbell's V-8 Juice Agar). Multiple hyphal tip subcultures from each root section subculture allowed the detection of the presence of multiple individuals and species of *Phytophthora* in individual soil samples.

Morphological identification

Prior to induction of sexual and asexual spores, the cultures were first examined for obvious morphological characteristics. Cultures were grouped using basic features and allocated to morphological groups (morpho-groups). This made it possible to deal with the large number of samples more effectively and to ensure that representatives of each group were later identified using molecular techniques.

Representatives of morpho-groups were induced to sporulate using a pond water wash (Drenth and Sendall 2001). The reproductive characters were then used to attempt identification using the key of Waterhouse (1963) and the Revised Tabular Key (Stamps *et al.* 1990).

Molecular identification

Investigating the population structure of *P. cinnamomi* requires both accurate identification of the species and a measure of genetic diversity. We investigated a molecular technique that could provide both a molecular identification and provide a measure of within-species genetic diversity. Initially, selected isolates were sent to the CRC-TPP for molecular identification. PCR technology is used to amplify part of the ribosomal DNA repeat region of *Phytophthora* that has been found to show little variation among isolates within a species but shows considerable variation between *Phytophthora* species (Drenth *et al.* 1999).

Once accurate molecular identifications were obtained, these isolates were used to identify RAPD (randomly amplified polymorphic DNA) primer amplification patterns to aid in the identification of other isolates. This process is an application of PCR (polymerase chain reaction), which allows extremely small quantities of DNA to be amplified into millions of copies in a short time. Random sequences of bases are used in the RAPD-PCR reaction that allows the genome of an individual to be randomly sampled for genetic variation (McDonald and McDermott 1993). The amplification products produced are separated using gel electrophoresis and can be visualised as bands of different molecular weights. The RAPD method is discussed in more detail in Section 1.4.

1.2.3 RESULTS

Morphological identification

Despite the use of a selective antibiotic medium, *Pythium*, which is closely related to *Phytophthora*, and two genera of true fungi, were also isolated from soil samples. However, *Phytophthora* could be distinguished by the following characters:

- hyaline (colourless) mycelium,
- white aerial hyphae (when in a mass) and
- diameter of the hyphae typically three (3) to eight (8) mm (depending on the age of the colony), (Waterhouse 1963).

Four morphological groups (morpho-groups) could be distinguished amongst the *Phytophthora* isolates obtained during this project. These were distinguished from one another by the characters listed in Table 1.1.

Table 1.1. A key to the morphological groups isolated from Mount Lewis and Tully Falls/ Koombooloomba Dam.

1b.	White aerial mycelium present	2
1a.	Little or no aerial mycelium present	3
2a.	Dense aerial mycelium on 10% V8 agar, aged hyphae coralloid, numerous typically botryose hyphal swellings, chlamydospores present, oospores absent in single culture.	**Group 1**
2b.	Sparse aerial mycelium on 10% V8 agar, dense in patches, no coralloid hyphae, oospores absent in single culture, papillate sporangium present in aerial mycelium.	**Group 2**
3a.	Abundant oospores in single culture on 10% V8 agar, aplerotic oospores, oogonia 28 μm diameter and tapering to base, spherical amphigynous antheridium, chlamydospores absent.	**Group 3**
3b.	None of the above combination of features.	**Group 4**

The Waterhouse key (1963) and the Revised Tabular key (Stamps et al. 1990) were used to attempt identification of the morpho-groups using the basic characters described in Table 1.1. The only morpho-group that could be identified tentatively to species was morpho-group 1, which was thought to be *P. cinnamomi*. This was based on the presence of only two characters, botryose hyphal swellings and coralloid mycelium (Table 1.2). The aerial mycelium character was not used in either the Waterhouse or the Revised Tabular keys (Table 1.2) All of the other morphological characters used to define the morpho-groups were found to occur in more than one species. Many of the characters cited by the keys, especially the sizes of particular structures, are reported as being extremely variable within the various species of *Phytophthora* (Stamps *et al.* 1990), or present in more than one species.

It was not possible to identify the species in all of the morpho-groups based only the characters in Table 1.1 or by using the reproductive structures that are traditionally used.

Molecular identification

Forty (40) isolates were identified using molecular techniques. One was contaminated during transportation and one other was identified as *Pythium*. This confirmed that a small group of isolates had been accurately identified as *Pythium* using morphological traditional characteristics. Of the remaining thirty-eight (38) isolates, ten (10) from morpho-group 1 were identified as *P. cinnamomi* and one (1) from morpho-group 2 as *P. palmivora* (Table 1.3) Seventeen (17) isolates from morpho-group 3 were identified as *P. heveae* and seven (7) as *P. katsurae*. Three isolates representing morpho-group 4 could not be identified using molecular markers (Table 1.3).

The RAPD molecular technique was useful in distinguishing between the species. Species-specific banding profiles were obtained using one primer (OPC-11), suggesting this primer may be useful in differentiating the species *P. palmivora*, *P. cinnamomi*, *P. heveae,* morpho-group 4 and Pythium (Figure 1.2).

Table 1.2. Morphological characteristics based on Waterhouse (1963) and Stamps et al. (1990). Charaters and states may be variably present in any isolate. Also included is sporangial papillation, often considered to be an important character: (P) character present, (l) character species specific.

Phytophthora species	Morphological Characteristics											
	Aerial mycelium	Chlamydospores present	Hyphal swellings on agar			Oogonia		Antheridia	Oospores	Sporangia		
			present	large, spherical, botryose	corraloid mycelium	tapered base	< 30 µm	spherical amphigynous	aplerotic	aerial on agar	papillate or semi-papillate	non-papillate
cactorum	n/a	P					P		P	P	P	
iranica	n/a	P					P			P	P	
pseudotsugae	n/a		P				P		P		P	
clandestina	n/a		P				P		P		P	
palmivora	n/a	P					P	P		P	P	
p. hetercystica	n/a	P								P	P	
megakarya	n/a	P				P	P			P	P	
arecae	n/a	P					P			P	P	
boehmeriae	n/a	P					P	P		P	P	
botryosa	n/a	P					P			P	P	
heveae	n/a					P	P	P	P	P	P	
katsurae	n/a	P				P	P	P	P		P	
meadii	n/a	P					P	P	P	P	P	
n. nicotianea	n/a	P	P				P	P	P	P	P	
n. parasitica	n/a	P					P	P	P	P	P	
capsici	n/a	P					P		P	P	P	
citrophora	n/a	P								P	P	
mexicana	n/a	P					P	P		P	P	
citricola	n/a	P					P			P	P	
syringae	n/a	P	P							P	P	
porri	n/a	P	P					P	P	P	P	
primulae	n/a	P					P				P	
colocasiae	n/a	P					P		P	P	P	
hibernalis	n/a						P				P	
ilicis	n/a					P	P				P	
infestans	n/a					P			P	P	P	
mirabilis	n/a					P	P			P	P	
phaseoli	n/a						P		P	P	P	
eriugena	n/a		P				P	P			P	
f. oryzobladis	n/a							P				P
m. megasperma	n/a	P							P			P
m. sojae	n/a	P										P
quininea	n/a	P	P						P			P
verrucosa	n/a						P		P			P
humicola	n/a		P				P		P			P
lateralis	n/a	P										P
insolita	n/a	P	P				P					P
cambivora	n/a		P			P						P
cinnamomi	n/a	P	P	●	P	P	P					P
cryptogea	n/a					P	P	P				P
drechsleri	n/a	P				P	P	P				P
cajani	n/a		P				P					P
melonis	n/a	P					P	P			P	P
sinensis	n/a		P				P	P				P
e. erythroseptica	n/a	P					P	P				P
e.pisi	n/a						P	P	P			P
richardiae	n/a		P				P					P
vignae	n/a		P				P	P	P			P
gonapodyides	n/a						P					P
japonica	n/a		P									P
undulata	n/a	P									P	P

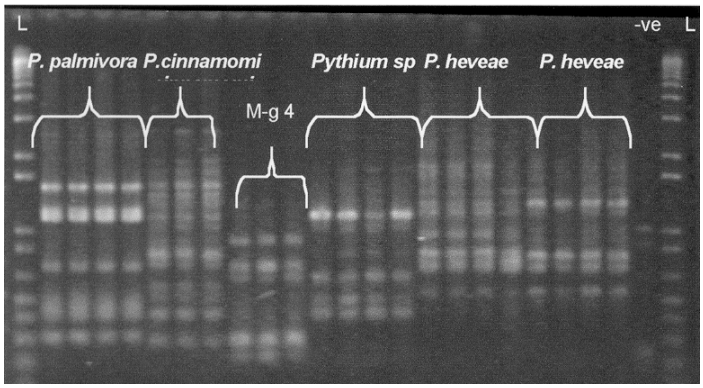

Figure 1.2. The RAPD profiles of Six individuals of different *Phytophthora* species produced by The random primer, OPC-11. Includes Includes *P. palmivora, P. cinnamomi, P. heveae,* morpho-group 4 (M-g 4) and *Pythium* sp., 3 TO 4 replicates per Individual.

Table 1.3. A comparison of the molecular and morphological identification performed on a select group of isolates from both Mount Lewis and Tully Falls.

Isolate Number	UQ Number	Morphological ID	Molecular ID
U2ML 4E	UQ 6247	Group 1	*P.cinnamomi*
A2ML2S	UQ 6270	Group 1	*P.cinnamomi*
U2ML 2N	UQ 6272	Group 1	*P.cinnamomi*
A6ML 1E	UQ 6275	Group 1	*P.cinnamomi*
U2ML 4E	UQ 6280	Group 1	*P.cinnamomi*
U2ML 4Nb)	UQ 6281	Group 1	*P.cinnamomi*
A2ML 3W	UQ 6282	Group 1	*P.cinnamomi*
A2ML 1N	UQ 6283	Group 1	*P.cinnamomi*
U2ML 1N	UQ 6284	Group 1	*P.cinnamomi*
U2ML 4Na)	UQ 6288	Group 1	*P.cinnamomi*
A3ML 2N	UQ 6278	Group 2	*P. palmivora*
A3ML 2Ea)	UQ 6234	Group 3	*P. heveae*
A4ML 4S	UQ 6235	Group 3	*P. heveae*
A3ML 1E	UQ 6236	Group 3	*P. heveae*
A3ML 2Eb)	UQ 6237	Group 3	*P. heveae*
C001 4Eb)	UQ 6238	Group 3	*P. heveae*
C001 4Ea)	UQ 6240	Group 3	*P. heveae*
A3ML 1W	UQ 6241	Group 3	*P. heveae*
U2ML 2Sb)	UQ 6242	Group 3	*P. heveae*
U2ML 1Eb)	UQ 6244	Group 3	*P. heveae*
A2ML 1E	UQ 6245	Group 3	*P. heveae*
A2ML 3Eb)	UQ 6246	Group 3	*P. heveae*
A2ML 3N	UQ 6248	Group 3	*P. heveae*
A2ML 1S	UQ 6249	Group 3	*P. heveae*
A2ML 4Eb)	UQ 6250	Group 3	*P. heveae*
A2ML 4Ea)	UQ 6251	Group 3	*P. heveae*
A2ML 3Ea)	UQ 6252	Group 3	*P. heveae*
U2ML 2W	UQ 6253	Group 3	*P. heveae*
U2ML 2Sb)	UQ 6271	Group 3	*P.katsurae*
A2ML 2N	UQ 6273	Group 3	*P.katsurae*
UIML 4Ea)	UQ 6274	Group 3	*P.katsurae*
U2ML 2Sc)	UQ 6277	Group 3	*P.katsurae*
A2ML 3Nc)	UQ 6279	Group 3	*P.katsurae*
U2ML 4W	UQ 6285	Group 3	*P.katsurae*
U2ML 1Ea)	UQ 6286	Group 3	*P.katsurae*
CK1ML	UQ 6239	Group 4	Unknown
A5ML Sapot.	UQ 6243	Group 4	Unknown
U2ML 2Sa)	UQ 6287	Group 4	Unknown

1.2.4 DISCUSSION

Dealing with large sample sizes such as those produced in distributional studies requires time efficient techniques for processing and identification of isolates. Initially, breaking down the isolates into more manageable morpho-groups using basic characters reduced the time required for subsequent molecular identification. The results of the molecular identification could then be compared with the morpho-groups relatively easily. Use of the RAPD molecular technique was an efficient and quick way of verifying the morphological identification of isolates.

The basic morphological characters that could be detected in pure culture were not sufficient for the identification of all isolates to species using Waterhouse's (1963) key or the Revised

Tabular key (Stamps *et al.* 1990). The primary character (aerial mycelium) that defined the morpho-groups used here was not used as a character in either of the keys, possibly because culture morphology can vary within species (Erwin & Ribeiro 1996).

Only one character (botryose hyphal swellings) was indicative as it was species-specific to *P. cinnamomi*. Although this character is only found in *P. cinnamomi* (Stamps *et al.* 1990), it may not always be expressed and in some cultures may be absent.

Although a tentative identification had been reached using morphology, the molecular results confirmed that isolates in morpho-group 1 were *P. cinnamomi*. While morphological identification failed to identify morpho-group 2 to species, the molecular technique was successful in identifying them as *P. palmivora* (Table 1.3). This was also the case for morpho-group 3, although the molecular results detected a further division within this group. Seventeen (17) isolates were identified as *P. heveae* and seven (7) as *P. katsurae*, despite all of those isolates being allocated to morpho-group 3 (Table 1.3). Morpho-group 4 could not be identified to species using either morphological or molecular techniques. Further investigation outside the scope of this project is required to determine whether morpho-group 4 isolates are a new species of *Phytophthora* or if they are a fungal genus with morphological characteristics similar to *Phytophthora*.

The RAPD molecular identification technique is a useful way of surveying large numbers of samples quickly, and has been shown to produce species-specific banding patterns. This technique has been used to produce a species-specific probe for *P. cinnamomi* without using the traditional and time consuming method of construction and screening of genomic DNA libraries (Dobrowolski and O'Brien 1993). While one of the aims of this study was to examine the level of genetic variation within populations of *P. cinnamomi*, this technique could also be used to detect differences between species of *Phytophthora*. One random primer (OPC-11) consistently showed a difference in banding pattern profiles between the species of *Phytophthora* analysed here (Figure 1.2).

The morphological technique of identification is inadequate for assessing *Phytophthora* species diversity at a site. The higher degree of resolution provided by molecular techniques provides a more reliable assessment of species composition from a site. Molecular identification gives more confidence to the initial tentative identification of morpho-group 1, which essentially was based on only one strong morphological character.

1.3 DISTRIBUTION OF *PHYTHOPHTHORA* IN TWO REGIONS OF FAR NORTH QUEENSLAND: MOUNT LEWIS AND TULLY FALLS / KOOMBOOLOOMBA DAM

1.3.1 INTRODUCTION

In previous assessments of *Phytophthora* distribution in the Wet Tropics, up to eleven species were identified (Brown in Gadek 1999). *P. cinnamomi* was found to be the most common species isolated from sampling sites, and more common in sites with obvious symptoms of canopy dieback than in healthy sites (Brown in Gadek 1999). On the other hand, Pryce (2000) found that the proportion of *P. cinnamomi* isolated from soils did not differ between affected and unaffected sites in the Tully Falls/Koombooloomba Dam area. Both studies relied on morphological characters for identification and may have underestimated the distribution and population structuring of *P. cinnamomi* in these areas.

This section examines the distribution and population structure of *Phytophthora* species, comparing the Mount Lewis and Tully Falls/Koombooloomba Dam areas. If the distribution and proportion of *Phytophthora* species differs significantly between the two regions, then each region needs to be considered independently. Therefore, the first hypothesis tested is that the distribution and population structure of *Phytophthora* species is not different between the Mount Lewis and Tully Falls/Koombooloomba areas. A second hypothesis – the level of inoculum of *P. cinnamomi* is not different between affected and unaffected sites – can be tested either by combining the regions if no difference is found, or within each region.

1.3.2 METHODS

Site selection

Mount Lewis and Tully Falls/Koombooloomba Dam are two of the four areas of Far North Queensland that have currently been mapped for canopy dieback and were the two regions chosen to be surveyed for *Phytophthora* in this study.

The affected sites at Mount Lewis were labelled using the following system:

- A = Affected sites, U = Unaffected sites
- The number of the site (1-7)
- The region in which the site was located: ML = Mount Lewis

i.e. MLA1 = Affected site one at Mount Lewis. In some cases, the codes are reversed, *i.e.* A1ML.

As Tully Falls has been studied previously, the labelling system previously used for those sites was maintained:

- P = Affected sites, C = Unaffected sites,
- The number of the site, (site 1 = 001)

i.e. P001 = Affected site one at Tully Falls (Pryce 2000).

The affected sites were paired with unaffected sites using the following criteria:

- located in the immediate vicinity (within 1 km) of the paired affected site,
- a similar distance from roads and watercourses and,
- on similar sloping ground, vegetation type and geology.

In the field at Mount Lewis a severely affected site was found close to MLA4 (MLA4b) and due to their close proximity only one unaffected site was used a comparison for both sites (MLU4). A third site was also sampled within the vicinity, which was intended as an unaffected site (MLA5). The number of sites that were sampled at Mount Lewis was seven (7) affected and five (5) unaffected sites (Figure 2.2) At Tully Falls/Koombooloomba Dam six affected and six unaffected sites were sampled (Figure 2.3 and Figure 2.4). The sites were located in the field using grid references obtained from the GIS database (Table 1.4).

Table 1.4. Grid references for affected and unaffected sites at Mount Lewis (approximate only) and Tully Falls/Koombooloomba. (all grid references use the Australian Geodetic Datum 1966)

Mount Lewis Grid References					
MLA1	318 307	8 165 290	**MLU1**	318 307	8 165 290
MLA2	317 250	8 165 300	**MLU2**	317 250	8 165 300
MLA3	317 700	8 164 650	**MLU3**	317 700	8 164 650
MLA4	316 800	8 163 820	**MLU4**	316 800	8 163 820
MLA4b	315 050	8 164 150		-	-
MLA5	316 963	8 164 251		-	-
MLA6	314 800	8 172 265	**MLU6**	314 800	8 172 265
Tully Falls Koombooloomba Dam Grid References					
PO01	353 025	8 022 175	**CO01**	353 325	8 022 675
PO02	354 764	8 016 574	**CO02**	354 845	8 016 900
PO03	346 893	8 033 443	**CO03**	346 543	8 033123
PO04	354 486	8 018 895	**CO04**	353 720	8 018 775
PO05	354 499	8 018 695	**CO05**	354 674	8 018 325
PO06	354 255	8 018 250	**CO06**	354 399	8 018 500

A somewhat different site selection strategy was used in the field at Mount Lewis in comparison with Tully Falls. At Mount Lewis a GPS was used to locate the point on the road nearest to the selected site. The GPS displayed the direction and distance to the selected site. From here, a compass was used to find the site.

As the mapped canopy dieback patches are relatively large (often several hectares in extent), only a small area within them could be sampled. Once in the general vicinity of the dieback patch, the area was scouted for the most severely affected area of the dieback patch. The purpose of the site selection strategy at Mount Lewis was to attempt to ground-truth the dieback patches. At Tully Falls the strategy was slightly different as this region has been sampled previously (Pryce 2000). The GPS co-ordinates were again used to locate the nearest point from the road, however, the exact direction and distance to the site was used by following compass readings. Using this method the majority of the sites that were used in the previous study were re-visited.

Soil sampling

The soil sampling protocol used in this study was modified from Pryce (2000) to ensure that comparisons could be made between the studies. Four ten (10) metre long transects radiating from the approximate central point of each site were measured with southerly, westerly, easterly and northerly bearings. At Mount Lewis four soil samples were taken from each transect at intervals of two and a half metres (2.5m). At Tully Falls/Koombooloomba Dam two samples were taken per transect at intervals of five metres (5m) from the central point. Soil samples were placed in clip-seal plastic bags and stored in a cool, dark place until ready for use.

To prevent cross-contamination between soil samples between each soil sample and each site the trowels were sterilised with five percent (5%) biodegradable bleach, then rinsed with water. Shoes were also sterilised in this way between sites. All of the bleach used for

sterilisation was collected and carried out with buckets and later disposed of appropriately. With the exception of flagging tape all sites were left as they were found. In total, two hundred and eighty (280) soil samples were collected from the two areas: one hundred and ninety-two (192) from Mount Lewis and eighty-eight (88) from Tully Falls. All soil samples were baited for *Phytophthora* using lupins and all root sections were plated, regardless of whether symptoms of infection were observed.

1.3.3 RESULTS

Sample design and limitations

Seven affected and five unaffected sites at Mount Lewis and six affected and unaffected sites at Tully Falls were sampled and treated as replicates for within and between the region comparisons. Affected and unaffected sites were paired, except for three affected sites at Mount Lewis, which were paired with just one unaffected site due to their close proximity to one other. Sixteen replicate soil samples were taken from each Mount Lewis site, and eight were taken from each site at Tully Falls/Koombooloomba.

The total number of samples that were positive for *Phytophthora* was one hundred and sixty-four (164): one hundred and seven (107) from Mount Lewis and fifty-seven (57) from Tully Falls. From the total number of samples taken from each region approximately half were positive for *Phytophthora*, slightly more for Tully Falls (Figure 1.3).

Phytophthora species diversity

Using a combination of morphological and molecular techniques, four *Phytophthora* species were identified from soil samples at Mount Lewis including *P. cinnamomi*, *P. heveae*, *P. katsurae* and *P. palmivora* (see Section 1.2). Only *P. cinnamomi* and *P. heveae* were isolated at Tully Falls. *P. katsurae* may be present at Tully Falls as it was not possible to distinguish this species from *P. heveae* using morphology alone. As only a small number of *P. katsurae* in total were identified using molecular techniques, the two species *P. heveae* and *P. katsurae* were pooled together. An unknown species, possibly a new species of *Phytophthora*, or a true fungus with similar morphological characteristics to *Phytophthora*, was isolated from both regions.

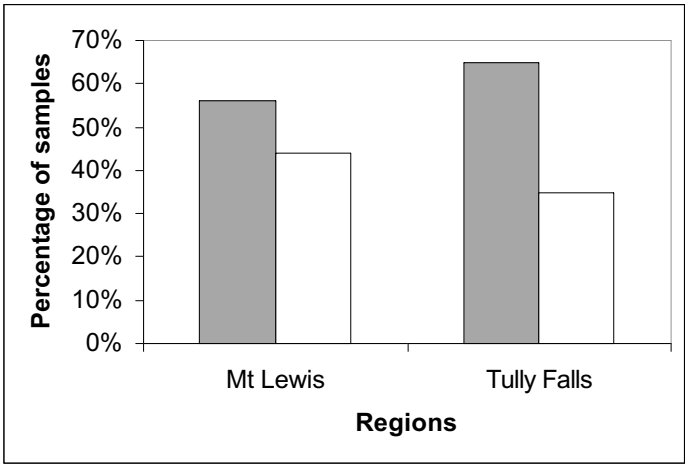

Figure 1.3. The percentage of soil samples that were positive (shaded) or negative (unshaded) for *Phytophthora* at Mt Lewis (n-192) and Tully Falls (n=88).

Region and canopy dieback interaction

Sites were treated as replicates, and the number of positive results from each site was used to generate the means used in the subsequent analyses. For the purposes of analysis, the isolate counts were divided into five groups:

(1) The number of *Phytophthora* isolations,
(2) The number of all *Phytophthora* isolates except *P. cinnamomi*,
(3) *P. cinnamomi*,
(4) *P. heveae*/*P. katsurae* and
(5) Morpho-group 4.

Two-way ANOVAs with Type III sums of squares, which corrects for unequal replication, were used to examine the effects of region and canopy dieback. This would determine

whether the mean number of positive results was different depending on where the samples were taken from, Mount Lewis/Tully Falls or affected/ unaffected sites.

A significant interaction was found between the two main effects, canopy dieback and region, for the total number of samples positive for *Phytophthora*, all species combined except *P. cinnamomi* and *P. heveae*/*P. katsurae* (Table 1.5).

Regional Effects: Mount Lewis and Tully Falls

The proportion of each species was calculated as a percentage of the total number of isolates obtained from each region. The most common was morpho-group 1 (*P. cinnamomi*), which made up 43% of the isolates from Mount Lewis and 39% at Tully Falls (Figure 1.4).

There was a significant difference between the regions for the total number of samples positive for *Phytophthora*, *P. cinnamomi*, and all species except *P. cinnamomi,* but not for *P. heveae*/*P. katsurae* (Table 1.5). This effect was only independent for *P. cinnamomi* (Table 1.5). There were more *P. cinnamomi* isolates detected at Mount Lewis than at Tully Falls.

Canopy dieback effects: affected and unaffected sites

Moving from the regional scale to the scale of canopy dieback patches, the distribution of *Phytophthora* species in affected and unaffected sites was examined.

Again the percentages were calculated using the total number of positive isolations for each species calculated as a proportion of the total number of positive isolates in each region. *P. cinnamomi* comprised 45% of isolates in affected sites compared to thirty-five percent (35%) in unaffected sites (Figure 1.7) Morpho group 4 was also found in higher proportion in affected sites (21%) than unaffected sites (14%). *P. palmivora* was only detected in affected sites, where it comprised 4% of isolates.

Table 1.5. The results of two-way ANOVAs for the total *Phytophthora* species, all species except *P. cinnamomi*, *P. cinnamomi*, *P. heveae*/*P. katsurae* and morpho-group 4 showing the F values, degrees of freedom and the significance for the treatments of region (Mount Lewis and Tully Falls), canopy dieback (affected and unaffected) and the interaction of the two main effects.

	F	df	P
Total *Phytophthora*			
region	85.757	1	**0.000**
canopy dieback	3.249	1	0.073
regionxcanopy dieback	8.725	1	**0.004**
All species except *P. cinnamomi*			
region	29.586	1	**0.000**
canopy dieback	0.073	1	0.787
regionxcanopy dieback	9.164	1	**0.003**
Total P. cinnamomi			
region	18.168	1	**0.000**
canopy dieback	9.832	1	**0.003**
regionxcanopy dieback	1.079	1	0.303
P. heveae/P. katsurae			
region	6.653	1	0.120
canopy dieback	0.197	1	0.659
regionxcanopy dieback	5.167	1	**0.027**
Morpho-group 4			
region	1.270	1	0.270
canopy dieback	2.786	1	0.107
regionxcanopy dieback	0.322	1	0.575

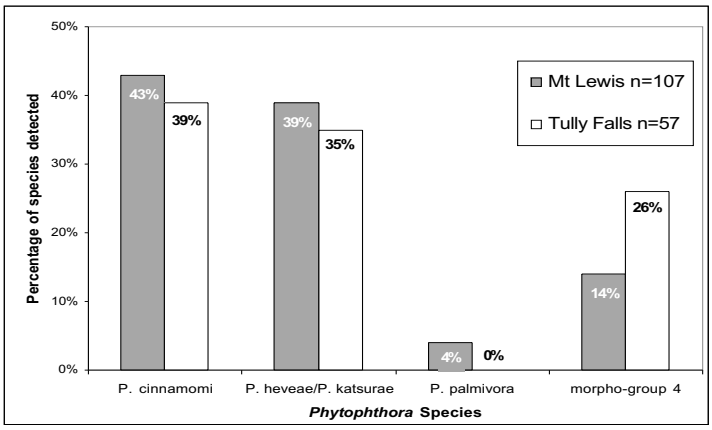

Figure 1.4. The percentage of *Phytophthora* species that were detected in the regions of Mount Lewis and Tully Falls.

The combination of *P. heveae* and *P. katsurae* showed a different relationship with canopy dieback, with a larger percentage (51%) found in unaffected sites than in affected sites (30%). The composition of *Phytophthora* species therefore appears to differ between affected and unaffected sites.

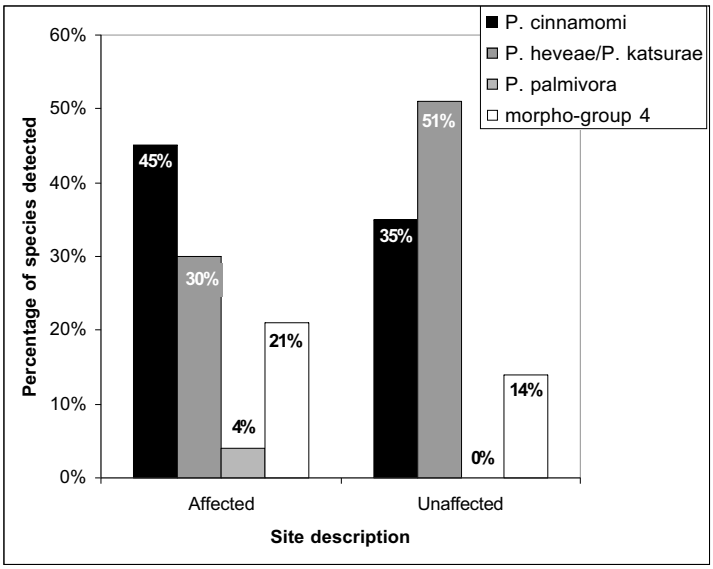

Figure 1.5. The percentage of *Phytophthora* species detected in sites affected (n=101) or unaffected (n=63) by canopy dieback

1.3.4 DISCUSSION

The most commonly isolated species was *P. cinnamomi,* followed by morpho-group 3 (*P. heveae* and *P. katsurae* combined). This finding supports those of Brown (in Gadek 1999) who found that out of 13,464 *Phytophthora* isolates, eighty-six percent (86%) were *P. cinnamomi* and the second most commonly isolated species was *P. heveae*. All other *Phytophthora* species were detected in extremely low proportions. Any differences in reported abundance between Brown and this project relate to a difference in the reliability of identification of isolates. As reported earlier, not all isolates can be positively identified using morphology alone.

As well as *P. cinnamomi; P. heveae*, *P. katsurae* and *P. palmivora* were detected at Mount Lewis, and at Tully Falls *P. heveae* and almost certainly *P. katsurae* were isolated. This is also in agreement with the findings of Brown (in Gadek 1999) who reported eleven different *Phytophthora* species in the rainforests of Far North Queensland. The only other study of *Phytophthora* in Tully Falls is that of Pryce (2000) who records only isolates that were identified using morphology as *P. cinnamomi.*

P. cinnamomi appears to be the most prevalent species at both Mount Lewis and Tully Falls/Koombooloomba. However, differences between the regions of Mount Lewis and Tully Falls do exist. More isolates were obtained from soil samples at Mount Lewis than at Tully Falls/Koombooloomga for all species except morpho-group 4 (Table 1.5). For all species except *P. cinnamomi* and the unknown species there were slightly more isolates found in unaffected sites at Tully Falls than in affected sites. In contrast there were always more isolates found in affected sites at Mount Lewis. Therefore, there was an interaction for all of the species except *P. cinnamomi.* The only species with significantly more isolates found in affected sites than in unaffected sites was *P. cinnamomi*. The level of inoculum of *P. cinnamomi* was greater at the scale of canopy dieback (in affected sites), as well as at the scale of region (Mount Lewis).

There were differences found between the regions. There were more isolates of all of the species detected at Mount Lewis than at Tully Falls. The likely factor that could have influenced the results may have been the different sampling strategies that were used between the regions. The sampling strategy at Mount Lewis was much more targetted than at Tully Falls. There is a level of error that is expected when grid references are obtained from a topographic map. The canopy dieback patches that have been found using aerial photography are large areas and samples are taken from only a small location within these patches. In the field at Tully Falls the grid references were strictly adhered to, the sites were

found using compass readings when the GPS failed to work under the rainforest canopy. At Mount Lewis the grid references were used to find the nearest point and direction to the canopy patches. The sites within the patches were then found by walking in and surveying the area for the most severely affected part of the dieback patch. While this was more time-consuming, it appears that it has had an important impact on the results that have been produced.

The more severely affected sites within the larger patches could be likened to the dieback fronts that are found in the ecosystems of Victoria and Western Australia (Weste *et al.* 1973). Within dieback fronts the population density of *P. cinnamomi* has been found to be extremely high (Weste *et al.* 1973). During the invasion phase, high frequencies of *P. cinnamomi* are found and this usually persists from one to three years after the initial infestation (Weste *et al.* 1973). The difficulty in isolating *P. cinnamomi* from old infestations has previously been noted by Dawson and Weste (1985).

1.4 GENETIC VARIATION OF *PHYTOPHTHORA CINNAMOMI* AT MOUNT LEWIS, FAR NORTH QUEENSLAND, IN RELATION TO CANOPY DIEBACK

1.4.1 INTRODUCTION

The genetic basis of virulence

P. cinnamomi is universally pathogenic; and is known to cause visible and dramatic symptoms of disease in well over 1000 species of plants (Erwin and Ribeiro 1996). Even in hosts that are thought to be resistant to *P. cinnamomi* infection, less visible symptoms may still be present (Weste and Marks 1987). There is some evidence that there may be virulent strains of *P. cinnamomi* that cause the most dramatic and visible effects on plants (Dudzinski *et al* 1993). For example, variation in the severity of disease was found when clones of *Eucalyptus marginata* were inoculated with a large number of different isolates of *P. cinnamomi* (Dudzinski *et al* 1993).

Virulence may have a genetic basis, as has been found for a number of different species of pathogens (Hermans *et al* 2000; Baele *et al* 2000; Smith and Stanosz 1995; de Wet *et al* 2000; Kolmer and Liu 2000). It is a possibility that specific genotypes (virulent strains) may occur within canopy dieback patches and could be responsible for the most pronounced disease symptoms (that is, complete canopy dieback) within localised sites.

RAPD analysis of genetic variation

The molecular technique chosen to detect the genetic variation between affected and unaffected sites is RAPD analysis (randomly amplified polymorphic DNA), which is an application of the PCR (polymerase chain reaction) (Williams *et al* 1990). Using this technique, a large number of isolates can be surveyed quickly using relatively small amounts of mycelium (McDonald and McDermott 1993). The RAPD technique is also especially useful for differentiating clonal lineages that are maintained by asexual reproduction (McDonald 1997), which has particular significance for investigations of *P. cinnamomi*.

RAPD markers have several drawbacks (particularly homology of bands) which limit their application in population genetic analyses (McDonald 1997). Despite these drawbacks, this technique is used because of its speed, relative cost effectiveness and suitability to broad surveys. Additionally, RAPD markers have been developed to allow species identification of isolates (see 1.2).

Aims

To examine the genetic variation of isolates from affected and unaffected sites at Mount Lewis. We wish to determine if genotypes of *P. cinnamomi* that occur in affected sites are significantly different from those that occur in unaffected sites.

1.4.2 METHODS

Growing isolates for DNA extractions

Since any soil sample may contain several individuals of any species, it is necessary to prepare pure cultures representing single individuals before any assessment of genetic variation can be undertaken.

The process involved in isolating pure *Phytophthora* cultures from soil samples has been explained in more detail in Sections 1.2 and 1.3.

Several different techniques for growing the isolates for DNA extraction were tested. The best was found to be one that has been previously used for growing *Sphaeropsis sapinea* (Smith and Stanosz 1995), and found to be simple and efficient. It involves taking a small sample of mycelium from a pure culture and using this to inoculate 0.5 mL of sterile 10% Campbell's V-8 Juice (clarified) in 1.5 mL Eppendorf tubes. After sufficient growth was observed the Eppendorf tubes were centrifuged at 13 000 rpm for 10 minutes. Centrifuging causes the mycelium to form a soft pellet and the V-8 juice can be aspirated leaving just the mycelium to use in the extraction.

DNA extraction

The DNA extraction method was that used for *Sphaeropsis sapinea*, developed by Gilbertson *et al.* (quoted in Smith and Stanosz 1995). Some minor modifications to this method were necessary. The mycelium pellets were washed twice with 10 mM Tris:1 mM EDTA. Lysis of the cell walls of the mycelium was performed by grinding in 250 μL of extraction buffer consisting of 100 mM Tris, 50 mM EDTA, 500 mM NaCl and 10 mM β-mercaptoethanol, using a Kontes Micropestle (Vineland, NJ). Another 250 μL of extraction buffer was then added along with 33 μL of 20% sodium dodecyl sulphate. This mixture was vortexed thoroughly then incubated in a water bath at 65° C for 10 minutes. To remove the cell wall debris 160 μL of 5 M potassium acetate was added, the mixture was briefly vortexed then centrifuged at 13 000 rpm for 10 minutes. After this step 450 μL of the supernatant was transferred to a new Eppendorf tube, leaving the cell wall debris pellet behind. The DNA was precipitated out of this supernatant by adding 225 μL of isopropanol, vortexing the mixture briefly then centrifuging at 13 000 rpm for 10 minutes. The supernatant was aspirated then the DNA pellet was washed in 500 μL of 70% ethanol and again centrifuged at 13 000 rpm for 10 minutes. The ethanol was aspirated and the DNA pellet was dried in a Speed-Vac until it had a glassy appearance (approximately 2 hours). The DNA was then dissolved in 100μL of sterile distilled water and left overnight to allow complete dissolution.

DNA concentration and purity were estimated using a spectrophotometer and concentrations were adjusted using sterile distilled water to 10ng/μL.

RAPD–PCR protocol

The RAPD-PCR protocol used is the standard technique followed in the Plant Systematics Laboratory, James Cook University Cairns Campus. It has been adapted from the *Taq* DNA Polymerase product protocol produced by Gibco BRL/Life Technologies. Several modifications were made to ensure that the protocol was optimised for *Phytophthora*, quantities of the chemicals were adjusted until the results were suitable. PCR amplifications were performed in 25 μL volumes in 0.5 mL microfuge tubes.

The master-mix solution for one reaction included the following:

- 15.15 μL of double sterilised distilled water
- 0.5 μL of 10 mM dNTPs
- 0.25 μL of 0.2% BSA
- 1 μL of 25 pmol/μL Oparon RAPD primer and
- 5 μL of template DNA.

The master-mix was covered with paraffin oil and centrifuged at 14 000 rpm for 1 minute to separate the oil from the solution. This was then soaked for 30 minutes at 60° C, which allows the BSA to bind to the protein contaminants resulting in a cleaner DNA template, using a Hybaid Omnigene Thermal Cycler. An enzyme mix was then added that consisted of:

- 2.5 μL of 10x magnesium deficient PCR buffer
- 0.5 μL of 50 mM magnesium chloride and
- 0.15 μL of *Taq* DNA polymerase.

A negative control was used with all of the same chemicals and only the DNA template replaced with sterile distilled water. Thermocycling was then performed with the Hybaid Omnigene Thermal cycler using the following program:

Step 1: 5 minutes at 95° C (initial denaturisation)
Step 2a: 60 secs at 95° C (denaturisation)
Step 2b: 60 secs at 36° C (primer annealing)
Step 2c: 2 min at 72° C (polymerisation and extension)
Step 3: Repeat steps 2a to 2c forty times
Step 4: 2 minutes at 72° C (final extension)
Step 5: cool to room temperature.

Gel electrophoresis

After the RAPD-PCR process had been completed, the amplification products were run out on a 1.5% agarose gel. Each well in the gel was loaded with 3 μL of loading dye and 10 μL of the PCR product. In at least three wells per gel, 4 μL of 0.1 ng/μL 1 kilobase ladder along with 3 μL of loading dye were loaded. The ladder was used as the standard to allow the sizes of the bands to be estimated. When the ladders were placed at either end and in the middle of the gel, their bands could be matched and allowed any distortion in the agarose gel to be corrected. The gel was then electrophoresed at 100 V and 400 amps for 10 minutes to move the products out of the wells, then run at 70 V and 400 amps for 180 minutes. After this time the gel was developed for 20 minutes in a dilute solution of ethidium bromide then rinsed in distilled water for 10 minutes. The gel was then photographed using a BIO RAD Gel Doc 1000.

Band scoring

The computer program, Biorad Quantity One was used to score the bands that were produced in the RAPD-PCR process. The lanes of the gel were framed with numbered lines and the bands were automatically detected and the sensitivity of detection was adjusted to 1.486. There were generally three ladders per gel, which were used as standards, their bands were numbered from 1 to 13 and lined up with each other using a function of the program. The detected bands for each lane were adjusted by examining the plotted band intensities, another function of the program. Any band with a smooth and distinct intensity peak was used and other bands without a peak were removed. The bands that were removed were generally shadows that appeared at the top or bottom of the gel. The match bands function was then selected using the lane with the most bands. The bands for all of the lanes were given numbers including the negative control lane. Any band that corresponded with a band in the negative control lane was not scored. All other bands were scored as 1 for present or 0 for absent in a matrix, for the band number that they had been matched as.

RAPD primer trials

Initial primer testing using four *Phytophthora* isolates that had been extracted was conducted. Identification to species had not been performed before the first primer trials. Two Operon primer kits were tested: Kit A and C, each with 20 random primers. The primers that produced polymorphic (multiple) bands were recorded.

Analysis of the RAPD matrix

The computer program Arlequin Version 2.000 (Schneider *et al.* 2000) was used to perform statistical analyses on the binary data that was produced from the band scoring process. This program measures and tests the standardised variance in allele frequency differences

among populations using an analogue of Wright's F_{st}., in an analysis of molecular variance (AMOVA), (Schneider *et al.* 2000). This test allows an indirect estimate of gene flow by analysing the allele frequency differences among pairs of populations. Tests can also be performed for hierarchical population structure among *a priori* specified population groupings.

The levels that were tested in this study included within populations (sites), between populations within groups (sites within affected and unaffected groupings) and between groups (affected and unaffected). All of the alleles and their frequencies are first combined and an assumption is made that the alleles have been sampled from one single randomly interbreeding population. The specific hierarchical levels are specified and sub-samples equivalent to those levels are taken. This is repeated for the number of permutations that are specified by the user, 10 000 in this study. The alleles and frequencies of the alleles that are sub-sampled are compared to the actual alleles and their frequencies that occur within the specified groupings. They are compared to determine whether the actual hierarchical population levels are significantly different from a randomly interbreeding population. The reason that this analysis is being used is because the affected and unaffected populations can be compared. This will allow the hypothesis that the individuals that occur in sites where there is a highly virulent outbreak of disease (affected sites) are genetically distinct from individuals that occur in unaffected sites to be tested.

1.4.3 RESULTS

RAPD primer trial results

There were 17 random primers found to be suitable for *Phytophthora*: OPA-02, 03, 04, 07, 08, 10, 11, 13, 18 and 19, and OPC-11, 12, 13, 14, 15, 16 and 18. When the RAPD-PCR technique was used for *P. cinnamomi* (isolates from morpho-group 1), 15 primers were found to be polymorphic: OPA-03, 07, 08, 09, 13, 18, and 19 and OPC- 11, (RAPD profile for this primer is shown in Figure 1.2) 12, 13, 14, 16, 18 (Figure 1.6), 19 and 20 and were subsequently used.

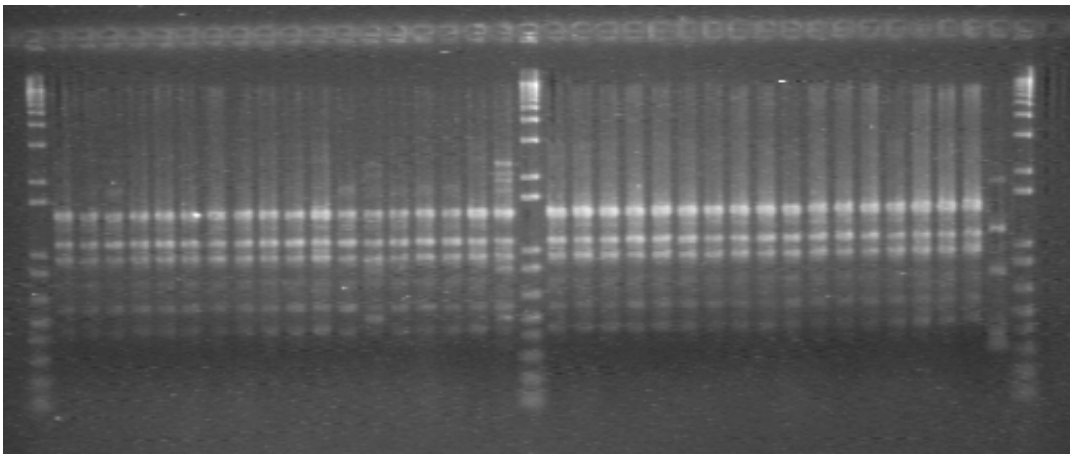

Figure 1.6 The RAPD profiles of 35 individuals of *P. cinnamomi* for one random primer (OPA-18), the similar (although not identical) band profiles support that all the individuals are the same species.

Sample size

DNA was extracted from thirty-five hyphal tip cultures belonging to morpho-group 1. These were from six affected sites and three unaffected sites at Mount Lewis. Nine individuals were from affected site one at Mount Lewis (MLA1), one from MLA2, two from MLA4, five from MLA5, two from MLA6 and five from MLA4b. Three individuals were from unaffected site one at Mount Lewis (MLU1), four from MLU2 and four from MLU6.

Genetic variation between affected and unaffected sites

AMOVA results for the following population groupings:

- between affected and unaffected groups,
- between the populations (each site) within the affected and unaffected groups, and
- within the populations (sites),

showed that all sources of variation were significant (Table 1.6). There was a significant difference between the affected and unaffected groups with almost eleven percent of the observed variation due to between group differences (10.71%), (Table 1.6). There was also a significant difference between populations (sites) within the affected and unaffected groups, with these differences being responsible for almost six percent (5.83%) of the observed variation, (Table 1.6). Most of the observed genetic variation (83.46%) was found within sites, (Table 1.6) The level of significance within populations (P = 0.0002) was greater than that between affected and unaffected groups (P = 0.02475) and between populations within the affected and unaffected groups (P = 0.03901).

Table 1.6. The AMOVA results showing the percentage of variation, the F-value, the significance and associated error of the significance for the three different sources of the variation that were tested.

AMOVA TABLE				
Source of Variation	**% Variation**	**F value**	**P**	P+/-
Affected vs Unaffected Groups	10.71	0.10711	**0.02475**	0.0016
Between Populations Within Groups	5.83	0.06527	**0.03901**	0.0019
Within Populations	83.46	0.16539	**0.0002**	0.00014

Geographic structuring of *P. cinnamomi* populations

The migration rates between all pairs of populations were estimated using the pairwise population divergence estimates from the AMOVA. These values were plotted against geographic distance and a Mantel's correlation calculated to determine if there was any geographic structuring of the *P. cinnamomi* populations within the Mount Lewis area. This test may help to explain why there was significant variation at all levels tested using the AMOVA. There was no correlation found between migration rate and geographic distance between the populations (sites), as shown in Figure 7. There was no increase or decrease in the migration rate as distance between the sites increased (Figure 1.7).

When significantly different pairs of populations were examined, the migration rate appeared to decrease when the distance was greater than 100 metres (log [distance] = 2), except for one point, which showed an increase in migration rate above that distance (Figure 1.8). There was one significant pairwise difference at 100 metres (log [distance] = 2). However, the majority of the significant pairwise differences occurred at distances greater than 1000 metres (log [distance] = 3). All significant differences between affected

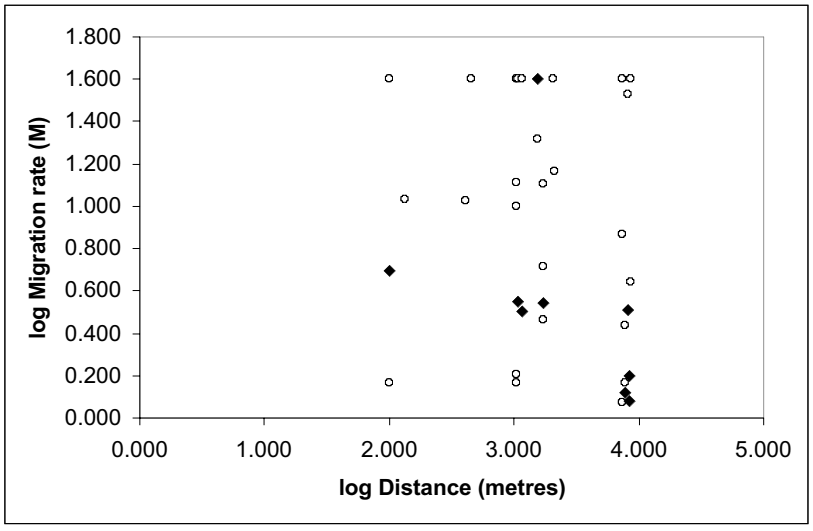

Figure 1.7. The log (migration rate) against the log (distance) between sites for *P. cinnamomi* at Mount Lewis. Symbols indicate pairwise divergence estimates that were not significant (o) and pairwise divergence estimates that were significant at *P* = 0.05 (♦).

and unaffected sites occurred at distances greater than 100 metres. No significant differences were observed between paired affected and unaffected sites.

Geographic structuring of affected and unaffected sites

A correlation between migration rate and distance was not found within affected or unaffected groups (Figure 1.8). The migration rate did not increase or decrease as distance increased when only the within affected and unaffected groups were examined (Figure 1.8). There does not appear to be a simple correlation between the distance and migration rate for the *P. cinnamomi* isolates that were sampled from Mount Lewis.

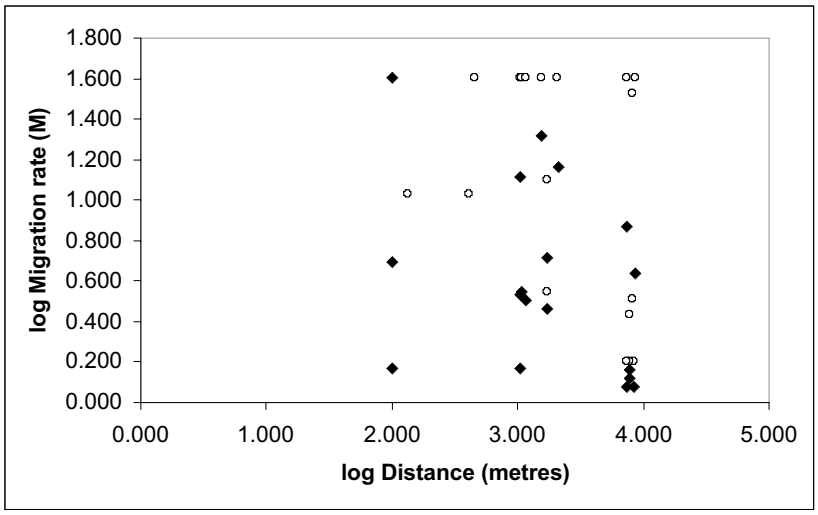

Figure 1.8. The log (migration rate) against the log (distance) between sites for *P. cinnamomi* at Mount Lewis. Symbols indicate within the affected and unaffected groups (o), and between the affected and unaffected groups (♦).

The significant differences between affected and unaffected groups, between the populations in the affected and unaffected site groups or within the populations do not appear to have a simple geographic explanation.

1.4.4 DISCUSSION

Unfortunately, not all *P. cinnamomi* isolates obtained from the Mount Lewis area could be used, as DNA extractions were sometimes unsuccessful. This affected sample size, and may have influenced the final results. This was especially true for one of the populations that had only one individual. There were three paired affected and unaffected sites. Three affected sites within the immediate vicinity of each other did not have a paired unaffected site for comparison. This over-representation of affected sites may also have influenced the results.

Although there were only 15 primers used in total, this appears to have been sufficient in detecting enough of the genetic variation that appears to exist in the populations of *P. cinnamomi* at Mount Lewis.

There was a significant difference between affected and unaffected groups. However, when this result was examined in more detail it appeared that the majority of the difference between groups could be explained by the geographic structuring of the populations. This is because the majority of the significant genetic variation between affected and unaffected sites appears to occur between unpaired affected and unaffected sites. The isolates in paired affected and unaffected sites were always found to have genotypes that were more similar to each other in comparison with other sites that were located at a distance greater than 1000 metres.

The inability to detect a correlation between migration rate and distance may also be the result of the low sample size. However, this could be an accurate depiction of what occurs within the populations of *P. cinnamomi*. There is no direct evidence that sexual reproduction

of *P. cinnamomi* occurs in nature although the two compatibility types may occur together (Dobrowolski 1999). The only genetic variation that has been detected elsewhere in Australia is three apparently clonally reproducing lineages, two of the A2 strain and one A1 strain (Dobrowolski 1999). If the isolates are clonally reproducing this may account for the inability to distinguish a correlation between migration rate and distance. The different lineages may occur together and the migration rate may be confused by the absence of sexual reproduction. Further investigation is required to determine whether the isolates used in this study correspond to the clonal lineages that have been found to occur in other regions of Australia.

Although there was a significant difference detected between affected and unaffected groups for the isolates of *P. cinnamomi* at Mount Lewis, most of that significance appears to occur between unpaired affected and unaffected sites. This would suggest possible geographic structuring of the populations. There was some structuring of the populations with significant differences detected for sites greater than 1000 metres apart. However there was no simple correlation found between migration rate and geographic distance. The relatively small sample size and using only three unaffected sites may have limited the strength of the analysis.

1.4.5 CONCLUSIONS

Verifying that *P. cinnamomi* does occur in a greater proportion within affected sites does not resolve the reason why pronounced levels of disease occur in patches within the rainforests of Far North Queensland. This species has also been detected under apparently healthy vegetation in all of the studies that have currently been performed in this region. The approach taken in this study to attempt to answer this question was to test the hypothesis that virulent outbreaks of disease may be caused by highly virulent strains of *P. cinnamomi*. When this possibility was examined it was found that there was a significant difference between the genotypes that occur in affected sites compared to unaffected sites within the region of Mount Lewis. However, a large proportion of this variation was found to occur between affected and unaffected sites that were more than 1000 metres apart. There was no significant difference found between paired affected and unaffected sites, which were located within 100 metres of each other.

A possible explanation is that there may have been geographic structuring of the populations that could explain why significant genetic variation only occurred at sites that were different locations. However there was no correlation found for migration rate and distance. Three clonal lineages of *P. cinnamomi* have been found to occur in Australia although this has not been tested in Far North Queensland (Dobrowolski 1999). This may account for the complex population structure that was found in this study. Clonal lineages may occur within the same locations and the absence of sexual reproduction may explain why the populations appeared to deviate from what would be expected for a randomly interbreeding population. The sample sizes and over representation of isolates from affected sites may have affected the results. Based on the results at hand, there does not appear to be a genetic explanation for the occurrence of canopy dieback patches in the rainforests of Far North Queensland. However, further investigation may be required before this possibility is completely discounted.

SECTION 2

IMPACT OF DIEBACK ON RAINFOREST CANOPIES

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2.1 INTRODUCTION

2.1.1 NATIONAL CONTEXT

Phytophthora cinnamomi is a fungus-like root-rotting pathogen with an extremely broad host range (Erwin & Ribeiro, 1996). It is soil-borne, and readily spread in soil or by surface or sub-surface water movement. It was first conclusively associated with dieback in jarrah in Western Australia in the mid 1960s (Podger, 1968), and since then its association with dieback has been widely reported in native ecosystems in southern and eastern Australia (Weste, 1994).

The disease that *P. cinnamomi* causes in susceptible species can lead to the death of the plant. If many species in a community are susceptible, removal of entire suites of species can lead to fundamental changes in ecology of systems, and threaten individual species with extinction (Weste, 1994). It therefore represents a significant threat to many native ecosystems. Its significance on a national scale has recently been recognised in its listing as a "Key Threatening Process" under the Commonwealth *Environment Protection and Biodiversity Conservation Act 1999*, and the development of the National Threat Abatement Plan (Environment Australia 2001).

2.1.2 REGIONAL CONTEXT

Patch death of rainforests was first observed in the 1970s in Far North Queensland at Dalrymple Heights on the Eungella Tablelands, west of Mackay, as well as in Garrawalt, to the west of Ingham (Brown, 1976). Extensive soil surveys were undertaken between 1975 and 1982 by Bruce Brown of the Queensland Department of Forestry (Brown, in Gadek 1999). He found almost 86% of the *Phytophthora* isolates from three different sites were *P. cinnamomi*. Whilst other *Phytophthora* species were present, they were found in relatively low proportions. In both the Garrawalt and Dalrymple Heights regions a higher proportion of *P. cinnamomi* was found in dieback-affected forest when compared to samples taken from apparently healthy vegetation (Brown, in Gadek 1999). An association between logged forest as well as the occurrence of feral pigs and higher percentages of *P. cinnamomi* isolations was also concluded (Brown, in Gadek 1999).

Brown's field investigations ceased in 1981, but patch death did not go away. In 1997, partly in response to concerns from the ecotourism industry, a preliminary report was commissioned by the Cooperative Research Centre for Tropical Rainforest Ecology and Management (Gadek 1997), recommending a workshop of invited specialist researchers to provide expert guidance for further research. The workshop (Gadek 1999) provided a foundation for the establishment of the current research program.

In 1999, whilst working on interpretation of aerial photographs for the Wet Tropics Management Authority (WTMA), David and Peter Stanton identified and mapped similar canopy dieback patches in upland rainforests in the Tully Falls/Koombooloomba, Lamb Range, Kirrama and Mount Lewis areas (Figure 2.1). Studies by (Pryce, 2000) of these mapped dieback polygons in the Tully Falls/Koombooloomba area found *P. cinnamomi* in

soils in the area. However, the pathogen was not restricted to dieback patches, nor did it occur at higher proportions within the dieback-affected sites in comparison to unaffected sites. Affected sites could be correlated with soil type, elevation, slope, and land profile (Gadek *et al.* 2001).

Although impacts on vegetation were apparent, there was no correlation found between affected sites and the presence (or absence) of particular species of plants (Pryce, 2000). There was no evidence of any impacts on particular species or suites of species between affected and unaffected sites, except the presence or absence of secondary symptoms of infection respectively (Pryce, 2000). Pryce (2000) further determined that a minimum of 3-4 soil samples were necessary t satisfactorily ascertain the presence of the pathogen in the rainforests of the Koombooloomba area.

This section presents findings of research undertaken to address the following specific recommendation arising from the 1998-2000 studies (Gadek *et al.* 2001), namely:

> *2. Continue to undertake field surveys to better characterise and ecologically describe the observed photopatterns including an assessment of the ecological health, successional status and identification of susceptible and resistant species.*

2.2 METHODS

2.2.1 DEFINITIONS

Mapped dieback polygon

These are defined in Gadek (1999) as “patch-death sites” - patches of apparent canopy death or thinning, identified from aerial photography, and transferred to a GIS layer. Size ranges (in the Tully Falls/Koombooloomba area) from one to ten hectares. Mapped dieback polygons (shown in Figure 2.1) do not define areas of uniform impact or effect. Rather, these boundaries delineate locations in which smaller patches of dead and dying canopies can be detected (D. Stanton *pers. comm.* in Gadek 1999).

Study site

Affected study sites were located within mapped dieback polygons, usually as close as possible to a dieback patch. Unaffected study sites were located in nearby forest areas outside the mapped dieback polygon, with similar physical characteristics (Gadek 1999).

Photopoint

Point at which a hemispherical photograph of the canopy was taken. Photopoints were laid out at random, at ten metre spacings on a north-south grid,. The first photopoint was always located as close to the centre of the study site as practicable. For assessments of floristics, each photopoint was assumed to be the central point of a 10 m × 10 m plot.

Hemiphot

Hemispherical canopy photograph.

Canopy openness

Calculated during hemiphot analysis: the ratio of canopy gaps and holes relative to the area of the whole hemisphere (Whitmore *et al.* 1993).

Compass bearings and navigation

In all field work, magnetic north was used as an approximation of grid north. Magnetic north lies approximately 7.0° east of grid north.

Datum
All grid references are with respect to the Australian Geodetic Datum 1966.

2.2.2 SITE SELECTION

Identifying study sites
The mapping of Stanton and Stanton indicated the presence of patch death in four areas of upland rainforest in the Wet Tropics World Heritage Area *viz:* the southern end of the Carbine Tableland around Mount Lewis, the Lamb Range north of Lake Tinaroo, around Tully Falls/Koombooloomba and near Kirrama west of Cardwell (Figure 2.1). Other potential dieback areas have been brought to our attention during the course of this research, and opportunistic observations of dieback were made during recreational ventures into the World Heritage Area.

Mapped dieback polygons were selected for study on the following criteria:
- they were greater than 200 m across (*i.e.* large and less likely to be missed due to navigational error)
- sites used in the studies of (Pryce, 2000) were re-identified where possible, and incorporated into this project
- they were within 500 m of vehicular access road

Locating study sites
Site grid references were determined from available mapping, and entered into a 12-Channel Garmin GPS receiver. Although rarely of use beneath the closed canopy of the forest, the GPS facilitated navigation to the point on the road closest to the selected dieback patch, from which point the direction and distance of the desired co-ordinates were displayed. It was necessary to navigate to the co-ordinates at the centre of the mapped dieback patch by dead reckoning. Once a dieback patch was identified, its centre (generally the point with the greatest amount of dieback visible overhead) was permanently marked for future reference. On clear dry days, the GPS could occasionally pick up sufficient satellites beneath the forest canopy to confirm the grid co-ordinates of the selected study site. In some areas, clearly defined dieback patches could not be identified within mapped dieback polygons. Opportunistic observations were made of dieback during bushwalking ventures.

Selection of the control, or unaffected sites followed the methods described in of Gadek (1999), p25.

Tasks undertaken at study sites
Time and financial constraints did not allow the full spectrum of assessments to be conducted at all sites in all areas, therefore, one affected and two or more unaffected sites were selected for detailed assessments, see Table 2.1.

Tasks undertaken at each study site are summarised in Table 2.1.

Table 2.1. List of study sites, and the research tasks carried out at each site. The methods employed in carrying out the respective search tasks are described in the sections indicated.

Study Site		Task				
		Baiting of Soils (Sect. 1.2)	Site Description (Sect. 2.2.2 & 2.2.3)	Soil Profile (Sect. 2.2.3)	Hemiphots (Sect. 2.2.4)	Canopy Composition (Sect. 2.2.5)
Koombooloomba/ Tully Falls	CO01	✓	✓	✓	×	×
	CO02	✓	✓	✓	×	×
	CO03	✓	✓	✓	✓	✓
	CO04	✓	✓	✓	✓	✓
	CO05	✓	✓	✓	×	×
	CO06	✓	✓	✓	×	×
	PO01	✓	✓	✓	×	×
	PO02	✓	✓	✓	×	×
	PO03	✓	✓	✓	✓	✓
	PO04	✓	✓	✓	×	×
	JP04	×	✓	×	✓	✓
	PO05	✓	✓	✓	✓	✓
	PO06	✓	✓	✓	✓	✓
Lamb Rg.	LRU1	×	✓	✓	✓	✓
	LRA1	×	✓	✓	✓	✓
	LRA2	×	✓	✓	✓	✓
Mount Lewis	MLU1	✓	✓	✓	×	×
	MLU2	✓	×	×	×	×
	MLU3	✓	×	✓	×	×
	MLU4	✓	✓	✓	×	×
	MLU6	✓	✓	×	×	×
	MLA1	✓	✓	✓	✓	✓
	MLA2	✓	✓	✓	✓	✓
	MLA3	✓	×	✓	×	×
	MLA4	✓	✓	✓	×	×
	MLA4 b	✓	×	×	✓	✓
	MLA5	✓	✓	×	×	×
	MLA6	✓	✓	×	×	×
	MLA14	×	✓	✓	✓	✓
Kirrama	KIU1	✓	✓	✓	×	×
	KIA1	✓	✓	✓	×	×
	KIA2	✓	✓	✓	×	×
Others	MMA1	✓	✓	×	×	×

2.2.3 SITE DESCRIPTION

Physical Description

For each site ticked in the task list (Table 2.1), the following physical features were noted:

- grid references
- substrate (parent material)
- topsoil description
- landform
- disturbance (natural and anthropogenic)
- forest structure and physiognomy
- list of plant species present

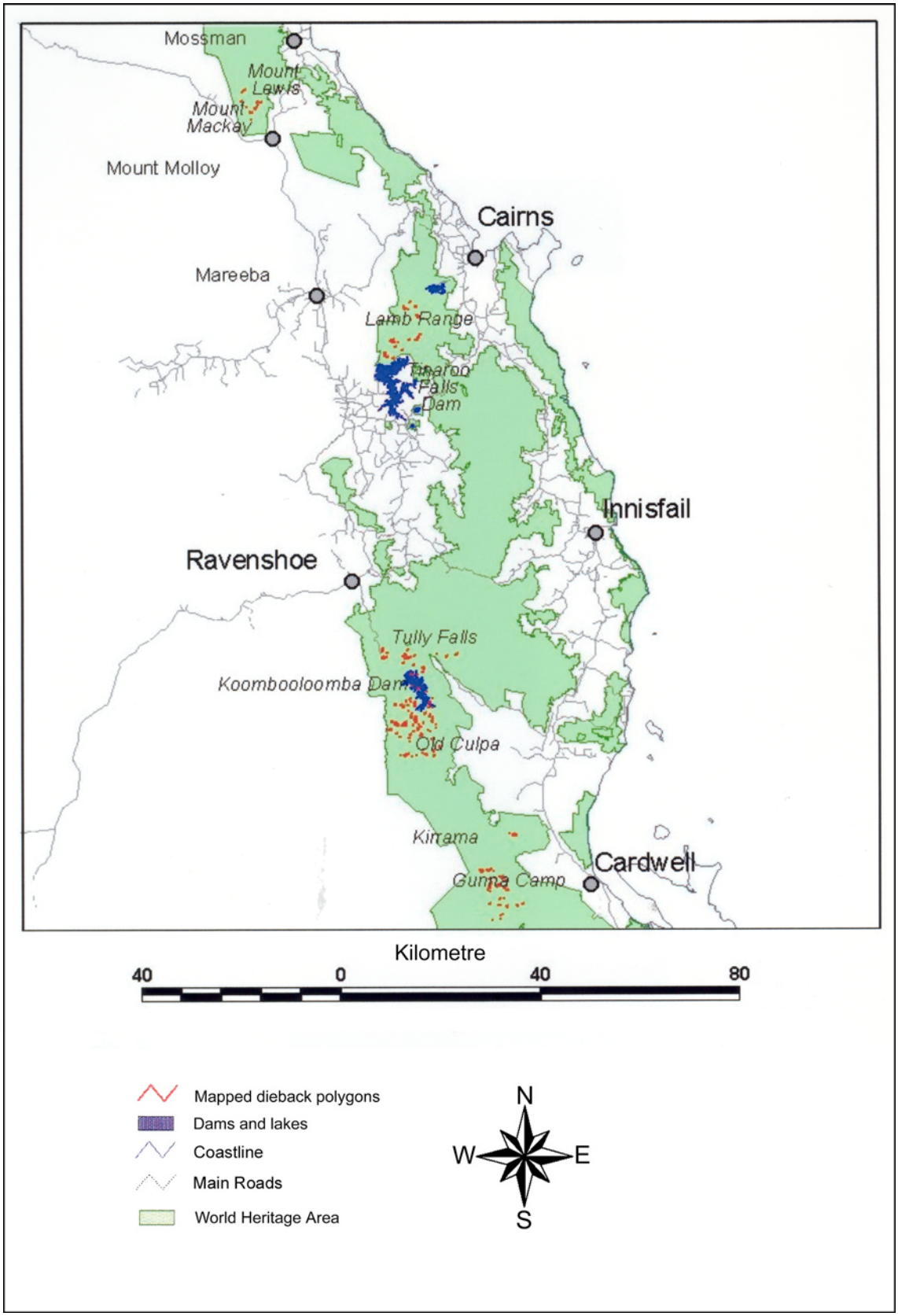


Figure 2.1. Mapped dieback polygons in the Wet Tropics World Heritage Area.

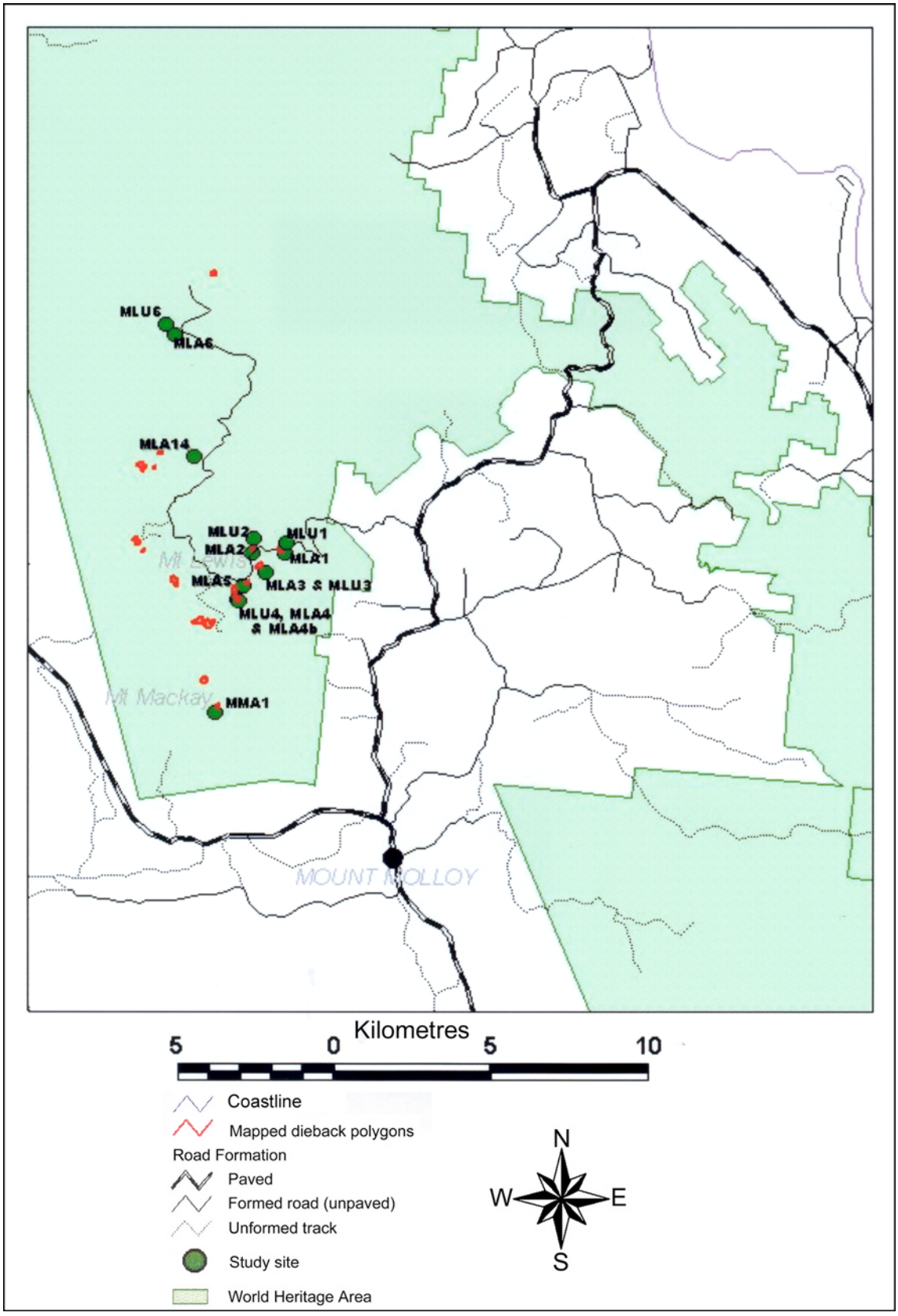


Figure 2.2. Study site locations in the Mount Lewis area.

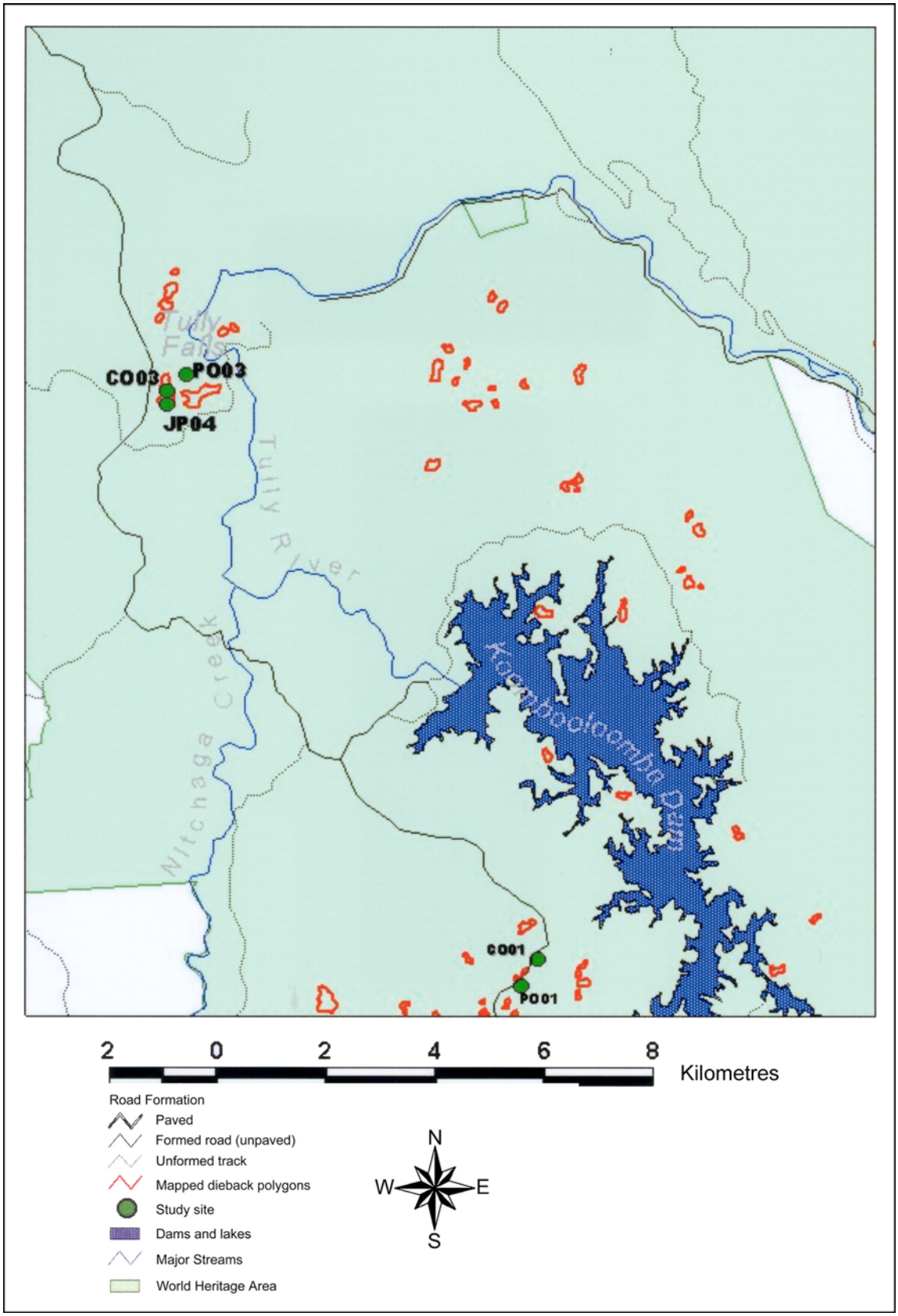


Figure 2.3. Study site locations in the Tully Falls area, north of Koombooloomba Dam.

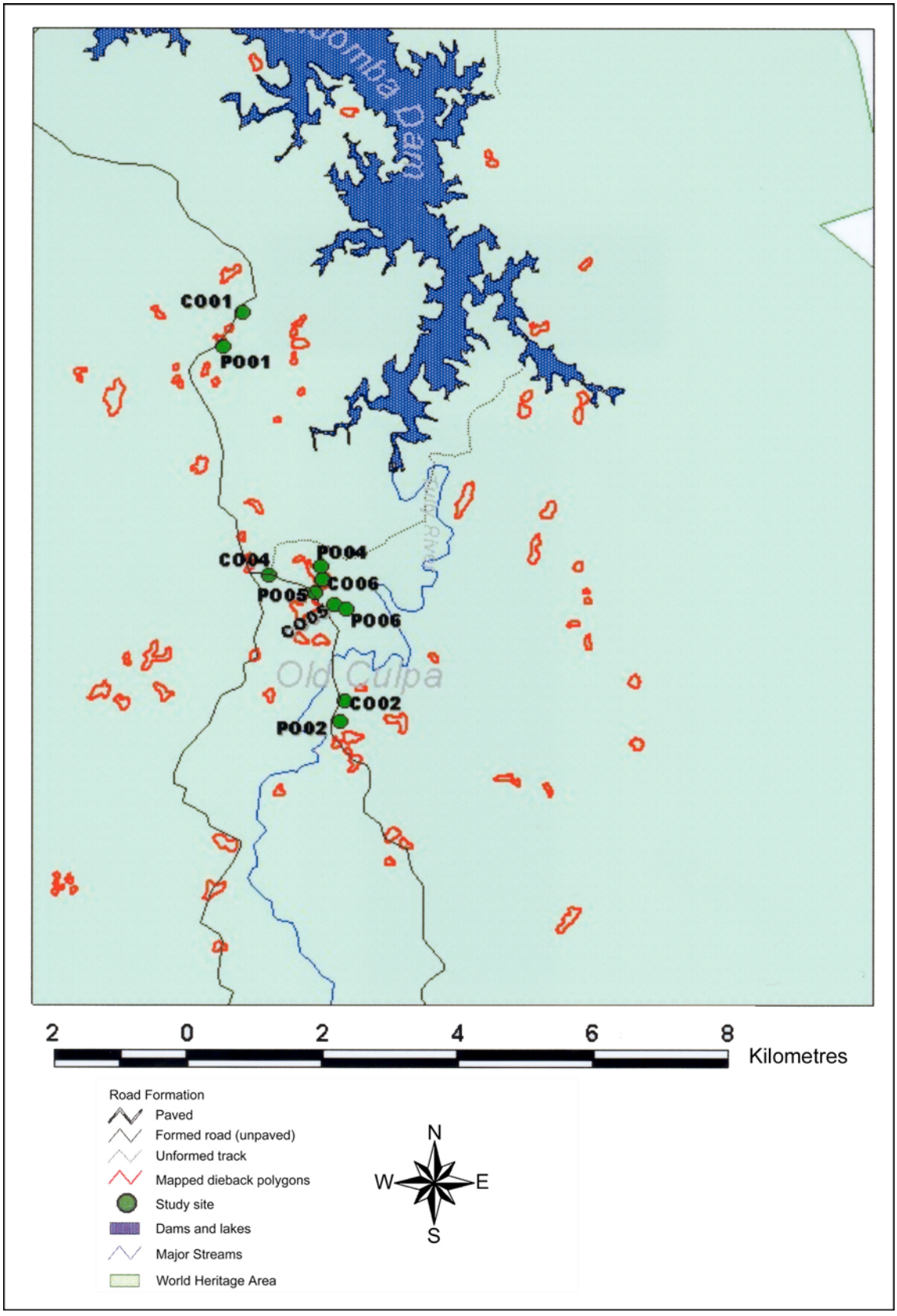


Figure 2.4. Study site locations south and west of Koombooloomba Dam.

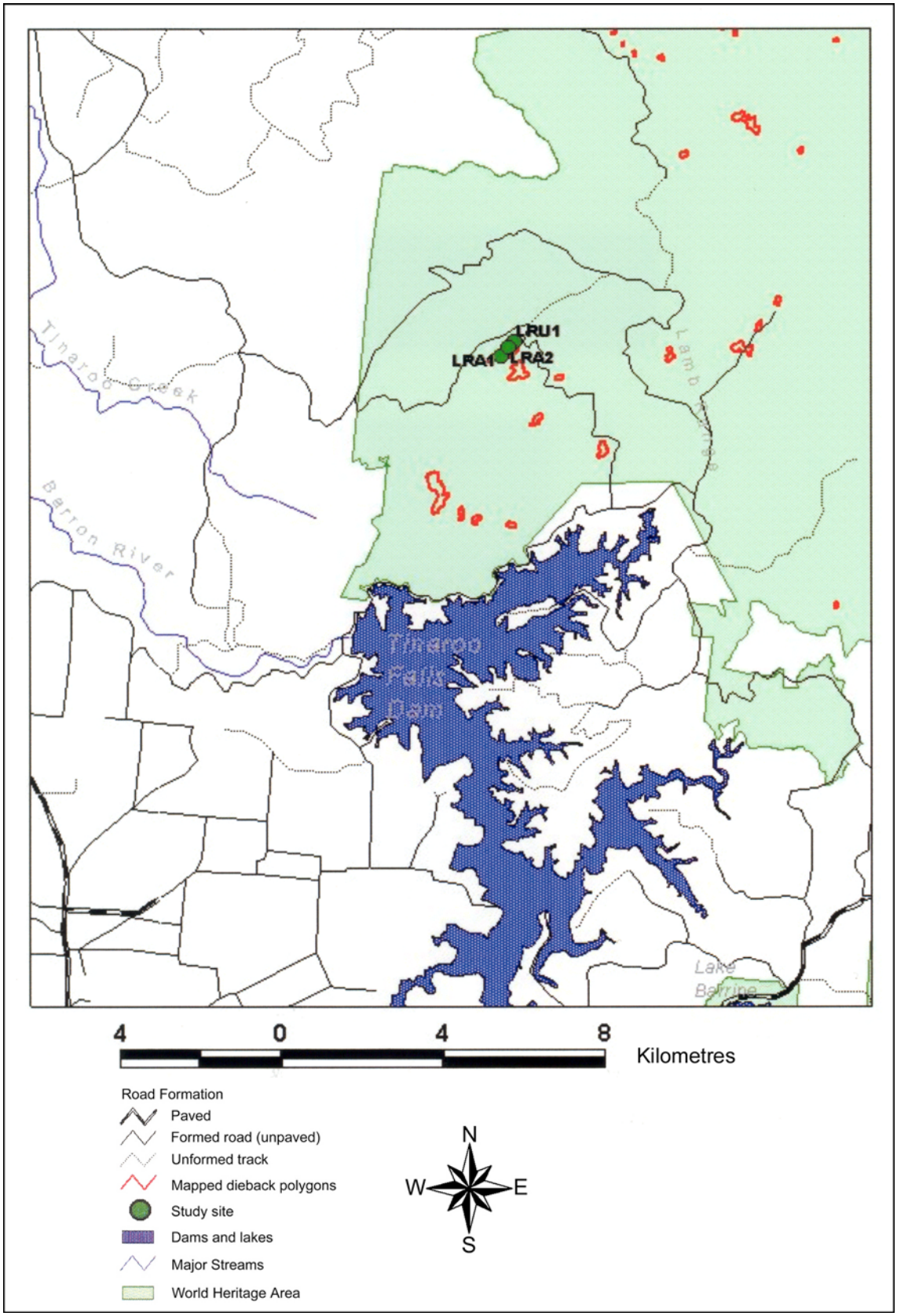


Figure 2.5. Study site locations in the Lamb Range area.

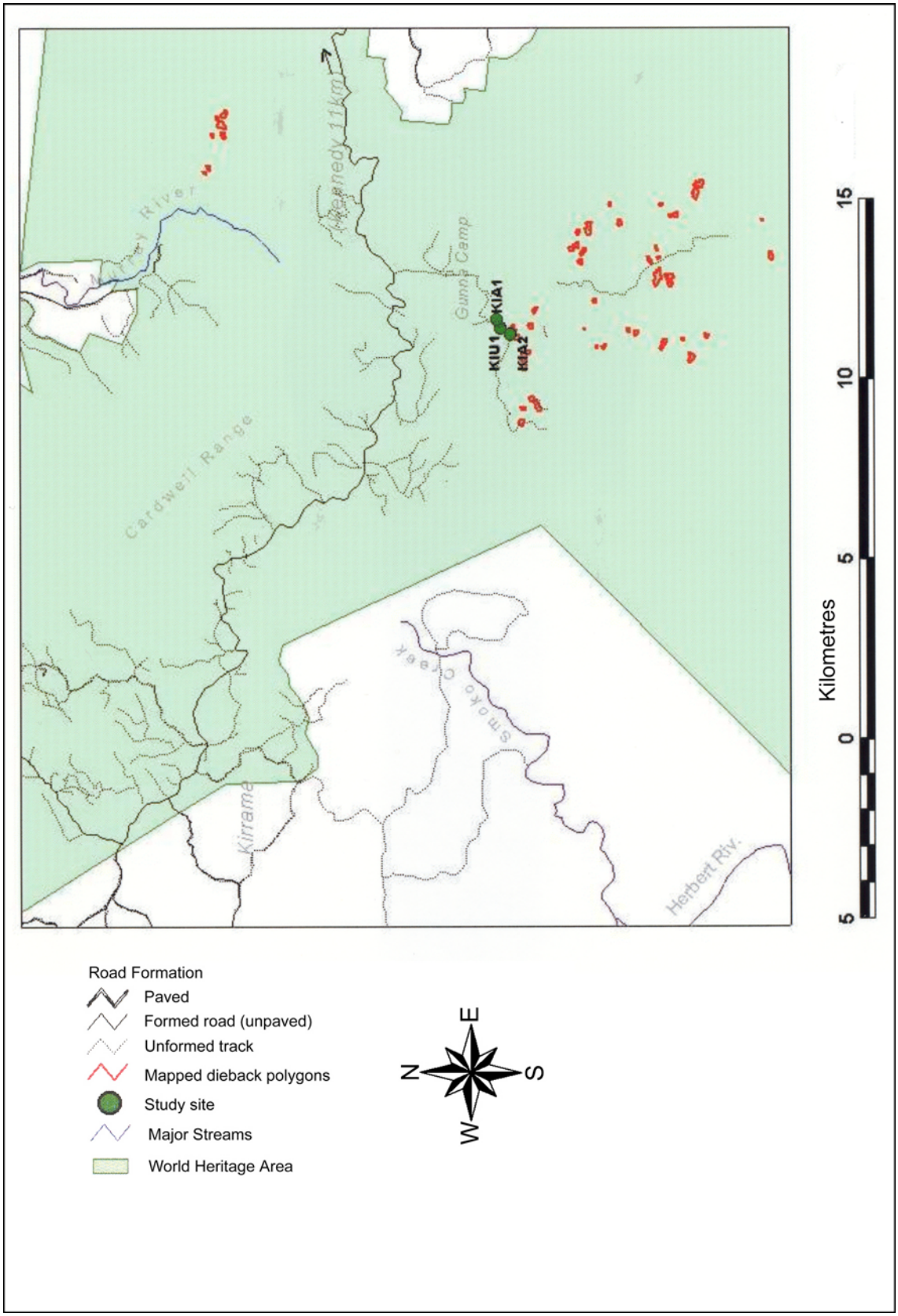


Figure 2.6. Study site locations in the Kirrama area.

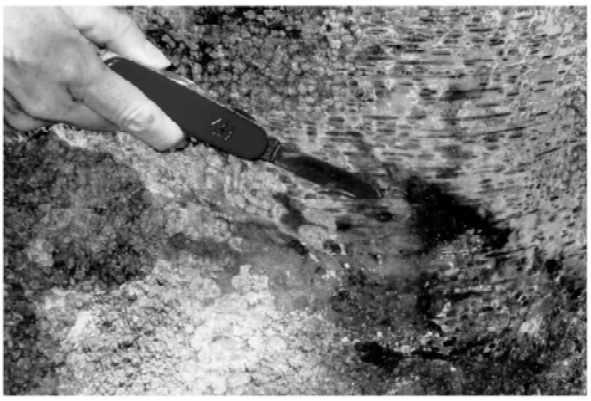


Figure 2.7. Lesion (darkened tissue) on lower trunk of Flindersia boujotiana, at site MLA4b, October 2001.

Figure 2.8. Recent dieback in trees near Mount Bartle Frere's Western Summit, December 2002.

Figure 2.9. Dieback-affected canopy trees at the Pilot Study Site (JP04), near Tully Falls, November 2001.

Figure 2.10. Severe dieback on steep hillsides near Mount Mackay, Carbine Tablelands, June 2002.

Forest Structure

The forest structure was described, and the Webb and Tracey (1975) forest type determined. The vegetation maps of Webb and Tracey (1975) were used to check type determinations. The corresponding Regional Ecosystem (RE: Sattler and Williams 1999) was identified, and the conservation status of the RE was ascertained with reference to the Queensland *Vegetation Management Regulation 2000*.

The biodiversity status of the RE, (as defined in Regional Ecosystem Description Database, EPA 2002) which reflects site condition, is also presented at URL:
http://www.env.qld.gov.au/environment/science/herbarium/regional_ecosystems

Site Health

The health of each study site, within a twenty metre radius of the central point, was visually assessed and rated using a modification of the method of Gadek *et al.* (2001), p27. The ratings used are defined as follows:

SEVERITY OF DIEBACK
0 – Insignificant with very few plants showing signs of leaf chlorosis or branch death
1 – slight (several plants as above)
2 – moderate (many plants as above, or some plants apparently dead or dying)
3 – severe (many dead, or gaps indicating loss of vegetation)

TREE FALL DAMAGE
1 – none
2 – slight (affecting small area in site only)
3 – moderate
4 – severe (affecting most of the site)

SITE DRAINAGE
1 – poorly drained
2 – well drained
3 – rapidly drained

DISTURBANCE TO SOIL SURFACE
0 – insignificant
1 – minor (small area patch-death)
2 – moderate (moderate or severe digging in small area)
3 – severe (most of site)

Soil Surveys

Soil assessments were aimed at addressing the hypothesis:

> Hypothesis 1*: Dieback sites exhibit a soil type where there is a greater level of change towards heavy clay than at unaffected sites.*

At the central point of each study site, profiles were dug with an auger up to 1.1 m deep, or shallower if rocks or ground water were encountered. For each horizon, field texture was assessed using the method of (McDonald and Isbell, 1990), and the number of field texture grade changes was compared between study sites. pH for the surface (A) horizon was measured using a 1:5 dilution of soil in distilled water (Isbell 1996).

Sites in the Kirrama area were the last sites set down for detailed study in the course of this study. In the light of soil observations made in the Koombooloomba area, it was decided to investigate soils at a greater intensity than had previously been done. Once suitable dieback sites were identified, five profiles were dug at each site, and the field texture of each horizon was recorded.

The five profiles were dug in a $\lesssim$ pattern aligned parallel to the slope of the hill, at five metre spacings.

2.2.4 CANOPY ASSESSMENTS

Canopy Photography

Hemispherical photographs of the canopy have been widely used to interpret the light environments beneath tropical rainforest canopies, in particular for the quantification of photosynthetically active radiation (for example, Turton, 1991, Whitmore *et al.* 1993, Siegenthaler, 1999). The method involves attaching a fish-eye lens with a 160° field of view to a standard 35 mm SLR camera, and taking ground-based images of the forest canopy. The resulting images can be analysed in a number of ways, but basically give an indication of the amount of light penetrating the canopy. As dieback areas are, by definition, areas where the tree canopy has thinned, analysis of hemiphots was employed to quantify gap size and the amount of canopy thinning that has taken place. Specifically, the hypotheses addressed were:

> Hypothesis 2: *Sites within mapped dieback polygons have a higher average canopy openness than sites in unaffected forest.*
>
> Hypothesis 3: *Sites within mapped dieback polygons will have a greater frequency of canopy gaps than in unaffected forest.*
>
> Hypothesis 4*: Canopy gaps in sites within mapped dieback polygons will be larger than those in sites in unaffected forest.*

Pilot study

To determine the sample size required to adequately assess the mean canopy openness for a study site, a pilot study was carried out at a moderately severe dieback patch near Tully Falls. At this site, labelled JP04 in previous studies, a one hectare plot was marked out. One hundred photopoints were marked out at 10 m intervals on a N-S grid within the plot.
The percentage canopy openness was calculated for hemiphots taken at 41 of the 100 photopoints (see "Hemiphot analysis", below). Analyses of the variation in canopy openness present across the site indicated between 15 and 20 hemiphots were required to sample the variation that naturally occurred across an affected site. This result was assumed to apply to all other Simple Notophyll Vine Forests photographed in this study.

Selecting photopoints

Starting at the central point of each selected site, sixteen to twenty adjoining photopoints were marked, laid our at random on a 10 m grid aligned to magnetic north. At some photopoints, wait-a-while (*Calamus spp.*: Arecaceae) was common to abundant. Where fronds of this vine, or any understorey shrubs, obscured the view of the canopy, they were removed.

Setting up the camera

Following the method of Turton and Duff (1992) hemiphots were taken with an Olympus OM1 camera body fitted with a Sigma 8 mm f1.4 fisheye lens. Fuji Neopan ISO 400 black and white film was used for all photographs. At each photopoint, the tripod was set so the plane of the film was 1.5 m above ground level. The lens was pointed skyward, and the camera body levelled with a bulls-eye spirit level. The top of the film was aligned to magnetic north.

Taking the photos

To avoid glare, images were recorded in periods of diffuse sunlight, usually before 0800 or after 1600, (Whitmore *et al.* 1993), under varying cloud conditions. Occasionally, heavily overcast conditions would permit a slight extension of the shooting time. Exposure time was set to 1/60 of a second, and three frames were shot – usually at aperture settings of 8, 11 and 16. When ambient light conditions were low, an additional frame was shot with the

aperture set to 4. Conversely, as light conditions increased, an additional frame was shot with the aperture set to 22.

The camera operator must take care to duck below the level of the camera when taking photographs with a fisheye lens.

Hemiphot analysis

Processed negatives were scanned direct to CD as 600 – 800 kilobyte JPEG image files by the processer. One hemiphot from each photopoint was selected for analysis, preference being given to photos with high contrast and low glare. Contrast and brightness were adjusted using Paint Shop Pro Version 5.01 (1998: Jasc Software and Access Softek) so that the images had a very high contrast and unexposed black background had a consistent average luminance value in all hemiphots. Images were reduced in size to 920 × 616 pixels and converted to 256 colour black and white bitmaps.

Canopy openness and gap size (if present) of images from each photopoint was evaluated using Winphot Version 5.0 (ter Steege 1996).

2.2.5 FLORISTICS AND CANOPY COMPOSITION

Many sites utilised in the studies of Pryce (2000) were revisited for this project, however the methods of floristic assessment applied in that project were not used. The objectives of this project required that floristic assessments focus on canopy composition.

Each photopoint was treated as the centre of a 10 m × 10 m plot. At each photopoint, trees likely to contribute to the canopy visible in the hemiphot were identified. Species names follow the system of Henderson (2002) unless otherwise specified. Size and health of each canopy tree were rated as follows:

SIZE
1 – less than 10 cm dbh
2 – 10–30 cm dbh
3 – 30–50 cm dbh
4 – greater than 50 cm dbh

Size classes were used as they allowed for a more rapid assessment of tree size than actually measuring the dbh. For calculations of stem basal area at each site, the $\Sigma(\text{size class})^2$ was assumed to be directly proportional to the actual basal area.

Shannon-Wiener diversity indices (Kent and Coker 1994) were calculated using canopy species lists, with $\Sigma(\text{size class})^2$ being utilised as a surrogate when estimating cover values.

HEALTH
1 – tree healthy
2 – some canopy thinning evident
3 – severe canopy thinning
4 – tree dead, but with leaves still present
5 – dead

Any tree with a health rating of >1 was considered “unhealthy” or affected by dieback. The symptoms used to define tree health ratings were consistent with those caused by *Phytophthora cinnamomi* infection, however, *Phytophthora* infection cannot be assumed in each and every unhealthy tree. Erwin and Ribeiro (1996) report that reddish-brown, resinous cankers were often visible on the trunks of infected hardwood trees, and these symptoms were sought as a confirmation of *Phytophthora* infection.

A rough map was prepared of the trees around each photopoint. This was used to identify large trees whose canopy impinged on more than one photopoint. In these situations, the

tree was noted as present above all photopoints, but for the purposes of data analyses was only recorded for the first photopoint above which it appeared.

To obtain an indication of the successional status of the community at each site, two measures were employed. The first was tree size (see above), the second was to assign a "successional status" code to each species identified in the surveys. Codes were derived from distribution and ecology notes provided in Hyland *et al.* (1999). The codes, and criteria used to assign the codes were:

Code	Criteria – The text of Hyland *et al.* (1999) describes the species as:	Examples
1	• favoured by disturbance • characteristic component of rainforest regrowth • regrowth species in disturbed rainforest	*Acronychia acidula* *Acacia sp.* (NQ BH 1344RFK) *(=A. celsa)* *Polyscias australiana*
2	• grows in a variety of rainforest types	*Tabernaemontana pandacaqui* *Rhodamnia sessiliflora*
3	• grows in well-developed rainforest	*Gillbeea adenopetala* *Buckinghamia celsissima* *Beilschmiedia tooram*
4	• grows in undisturbed rainforest	*Psychotria* sp. *(Utchee Ck H. Flecker NQNC5313)* Harpullia frutescens
No code	• Species not described in Hyland *et al.* (1999) • Successional status not clear or sufficiently specific in the distribution and ecology notes	*Most vines, ferns and orchids* *Baloghia inophylla* *Wilkiea angustifolia* *Alpinia arctiflora*

The hypotheses to be addressed by these surveys were:

Hypothesis 5: *Sites within mapped dieback polygons have lower stem basal areas than similar sites in unaffected forest.*

Hypothesis 6*: Species in susceptible taxonomic groups will be present at lower densities in mapped dieback polygons than in unaffected forest.*

Hypothesis 7: *Mapped dieback polygons will have lower living stem densities than unaffected forest.*

Hypothesis 8: *Mapped dieback polygons contain fewer taxa than unaffected forest sites* (Gadek *et al.* 2001).

Hypothesis 9: *Mapped dieback polygons are at an earlier successional stage (because of the impacts of dieback) than unaffected forest*.

Hypothesis 10: *Some taxa are restricted to unaffected (or affected) sites* (Gadek *et al.* 2001).

2.2.6 ANALYSES

Hypothesis 2: *Sites within mapped dieback polygons have a higher average canopy openness than sites in unaffected forest.*

Mean canopy openness (CO) values were calculated for each site. These values were normalised using the transformation: $\sin^{-1} \sqrt{(CO/100)}$. The transformed data were compared using one-way ANOVA, testing the null hypothesis that there is no difference in mean canopy openness between affected and unaffected forest.

Hypothesis 5: *Sites within mapped dieback polygons have lower stem basal areas than similar sites in unaffected forest.*

For comparisons of stem basal areas, (size class)^2 was utilised as a surrogate for actual basal area (see above). The mean stem basal area per photopoint was calculated for each study site. The site means were compared using one-way ANOVA, testing the null hypothesis that there is no difference in mean living stem basal area between affected and unaffected forest.

As a further comparison, the stem counts for each size class were pooled for affected and unaffected sites, and a χ^2 homogeneity test applied to test the null hypothesis that there was no difference between affected and unaffected sites in the proportion of stems belonging to each size class.

Hypothesis 6: *Species in susceptible taxonomic groups will be present at lower densities in mapped dieback polygons than in unaffected forest.*

Using pooled data from field studies, "field susceptible" species were arbitrarily designated as those species with five or more stems assessed on affected sites, and two or more of those stems being assigned a health rating of more than one. "Field resistant" were defined as species with five or more stems assessed on affected sites, and all of those stems healthy.

Stem counts for sites within an area were divided into three groups – "field susceptible", "field resistant" and "other" and a χ^2 homogeneity test applied to test the null hypothesis that proportions of stems in the three groups were the same across all sites. Each area was tested separately, and the combined data for affected and unaffected forest was compared.

Hypothesis 7: *Mapped dieback polygons will have lower living stem densities than unaffected forest.*

For comparisons of stem basal areas, the mean number of stems per photopoint was calculated for each study site. The site means were compared using one-way ANOVA, testing the null hypothesis that there is no difference in mean number of living stems between affected and unaffected forest.

Hypothesis 8: *Mapped dieback polygons contain fewer taxa than unaffected forest sites (Gadek et al. 2001).*

For comparisons of species diversity between sites in an area, only data from the canopy assessments of the first sixteen photopoints was utilised. For the Pilot Study site (Site JP04), data from sixteen randomly chosen adjoining photopoints were selected starting at the centre of the site. These data were used to determine:

a) the total number of species present at a site
b) the Shannon-Wiener diversity index (Kent and Coker 1995). In calculating this value, Σ(size class)^2 for a particular species was used in place of cover values.

One-way ANOVA was used to test the null hypotheses that there was no difference in gross numbers of species (or Shannon-Wiener diversity indices) between affected and unaffected forest.

Hypothesis 9: *Mapped dieback polygons are at an earlier successional stage (because of the impacts of dieback) than unaffected forest.*

The stem counts for each successional class were pooled for affected and unaffected sites, and a χ^2 homogeneity test applied to test the null hypothesis that there was no difference between affected and unaffected sites in the proportion of stems belonging to each successional class. Unclassified species were excluded from the analysis.

2.3 RESULTS

Site descriptions for the different study areas are presented separately. Other results, including hemiphot analyses and outcomes of floristic surveys and tree health assessments, are combined for presentation.

2.3.1 SITE DESCRIPTIONS, TULLY FALLS/KOOMBOOLOOMBA

Site descriptions for Tully Falls/Koombooloomba are presented in Table 2.2.. Study sites were mostly within 500 m of the Tully Falls-Culpa Road, spread over a north-south distance of 19 km. Study site localities are shown in Figure 2.3 and Figure 2.4.

Table 2.2. Physical description of sites in the Tully Falls/Koombooloomba area. Unless otherwise ntoed, these sites correspond to sites described in Gadek *et al.* (2001).

Site Code & Locality Name	Survey Date	Grid Co-ordinates	Altitude	Landform	Forest Type
CO01 Tully Falls – Culpa Road, Koomboo-loomba	15-Nov-01	353325, 8022675	860 m	Hillcrest & moderately inclined slope on rolling hills. Easterly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Notes: E-W Transect. Evidence of previous logging activities includes logging tracks and alien plant species on forest edges.				
CO02 Old Culpa, near Tully River	16-Mar-02	354845, 8016900	770 m	Gently inclined slope on undulating low hills. Southerly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Notes: Some evidence of logging activities. Culpa Road nearby. Canopy to 25m.				
CO03 Tully Falls	13-Nov-01	346543, 8033123	750 m	Gently inclined slope into a drainage depression amongst rolling low hills. Easterly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Notes: New site – site markers could not be found at 346725, 8033625. Prominent canopy species include *Acacia celsa* & *Cardwellia sublimis*				
CO04 Old Culpa	14-Nov-01	353720, 8018775	820 m	Moderately inclined hillslope on rolling hills. Easterly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Notes: N-S Transect. Evidence of previous logging activities. Forest to 35 m high. Prominent canopy species include *Flindersia pimenteliana, Syzygium kuranda, F. bourjotiana* & *Pouteria euphlebia.*				
CO05 Old Culpa	17-Mar-02	354674, 8018325	780 m	Gently inclined hillslope on undulating low hills. North-easterly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Notes: No evidence of anthropogenic disturbance. Forest to 25 m with occasional emergents to 30 m.				
CO06 Old Culpa	17-Mar-02	354399, 8018500	780 m	Very gently inclined hillslope on gently undulating rises. Southerly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Notes: Evidence of previous logging activities, although inconspicuous. *Cyathea rebeccae* prominent in under-storey. Forest to 25 m with occasional emergents to 30 m.				
PO01	16-Nov-01	353025, 8022175	860 m	Gently inclined slop on undulating low hills. North-westerly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Culpa Road nearby. Canopy to 25 m with occasional emergents. SE - NW Transect.				
PO02 Old Culpa	16-Mar-02	354764, 8016574 [1]	815 m	Gently inclined slope on undulating hills. East-south-easterly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Evidence of previous logging activities observed. *Cyathea rebeccae* and ground ferns prominent in understorey. Canopy height to 30m. New site – site markers could not be found at 354890, 8016540				
PO03 Tully Falls	13-Nov-01	346893, 8033443	760 m	Moderately inclined slope on rolling low hills. Northerly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Abundant *Dendrocnide morioides.* New site – site markers could not be found at 346520, 8033230 Prominent canopy species: *Cardwellia sublimis, Flindersia bourjotiana,* & *Gillbeea adenopetala*				

Site Code & Locality Name	Survey Date	Grid Co-ordinates	Altitude	Landform	Forest Type
PO04 Old Culpa	14-Nov-01	354486, 8018895	815 m	Gently inclined hillslope to footslope on undulating low hills. East-south-easterly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Dieback not particularly evident. Canopy height 25 – 30 m. Located 61 m to the east of the site described in Pryce (2000)				
JP04 Tully Falls	27-Jan-02	346544, 8033123	760 m	Gently inclined hillslope on undulating low hills. North-north-westerly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Pilot Study Site (Figure 2.8) Evidence of previous logging activities, including snig tracks and marked trees. *Calamus* sp. particularly abundant at this site. Canopy height to 30 m.				
PO05	16-Mar-02	354499, 8018695	780 m	Hill crest on gently undulating rises. South-easterly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	No evidence of anthropogenic disturbance. *Cyathea rebeccae* and ground ferns prominent in understorey. Some dieback evident. Canopy height to 30 m. Prominent canopy species: *Elaeocarpus sericopetalus, Flindersia bourjotiana* & *Cardwellia sublimis*				
PO06	16-Mar-02	354255, 8018250	800 m	Gently inclined hillslope on undulating low hills. North easterly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	No evidence of anthropogenic disturbance. Canopy height to 20m. Prominent canopy species: *Halfordia scleroxyla, Flindersia bourjotiana* & *Syzygium kuranda*				

Landform

The topography of study sites consists of undulating low hills with shallowly incised drainage lines. The area is situated on the northern Ingham Batholith (a suite of igneous rocks extending from the Tully Gorge to Kirrama [Bain and Draper 2000]). Rhyolites underlie northern study sites, granites underlie those further to the south. Altitude lay between 750 m and 860 m. More detailed landform descriptions are given in Table 2.2.

Forest Structure

The forest around each study site was classed as Type 8 (Simple Notophyll Vine Forest), as defined by Tracey and Webb (1975) which corresponds to Regional Ecosystem (RE) 7.12.16 (Sattler and Williams 1999)(Table 2.3). Some sites, such as PO03, displayed a degree of structural complexity that approached Type 5a (Complex Notophyll Vine Forest) – RE 7.8.4. Gadek *et al.* (2001) mapped the dieback polygons of Stanton and Stanton in the Tully Falls/Koombooloomba area with respect to geology and Tracey and Webb (1975) vegetation communities. These polygons fall within several forest types (Table 2.3). Corresponding REs area shown, and their conservation status (as listed in the Queensland *Vegetation Management Regulation 2000*) is given.

Site Health

Changes in site health during the period 2000-2002 are given in Table 2.4. In 2000, site health was assessed by Josephine Pryce and Will Edwards. The same assessments were made in 2002 by Stuart Worboys and Sandra Abell.

The severity of dieback at sites PO05 and PO06 increased over the two year interval between observations. Unaffected sites showed no change in dieback severity, except site CO02. Tree fall damage showed either no change or a reduction in severity at all sites. No evidence of pig damage or other soil disturbance was observed within any site in the Tully Falls/Koombooloomba area, although some pig damage was seen in a drainage line near JP04 – the Pilot Study site. *Phytophthora cinnamomi*, and other *Phytophthora* species, were identified from a majority of field study sites, but not from sites PO01 and PO02, supposedly dieback-affected sites. The numerical results of *P. cinnamomi* isolations cannot be validly compared, as both the sampling methodology and the species identification methods were changed. These results are discussed in more detail in Section 1.

Table 2.3. Regional Ecosystems containing mapped dieback polygons in the Tully Falls Koombooloomba area.

Regional Ecosystem	**Tracey & Webb (1975) Forest Type**	Description (Sattler and Williams 1999)	**Biodiversity Status**	**Conservation Status (VMA)**
7.8.1	**1a**	Complex mesophyll vine forest on very wet and wet lowlands and foothills, on krasnozem soils derived from basalts and basic volcanic parent material.	**Of Concern**	**Of Concern**
7.8.4	**5a**	**Complex notohyll vine forest on cloudy wet basalt uplands and highlands.**	**Of Concern**	**Of Concern**
7.12.14	13c	Notophyll vine forest with rose gum (*Eucalyptus grandis*) emergents on cloudy wet granite and rhyolite upland ridges.	Not of Concern	Not of Concern
7.12.15	13f	Notophyll rainforests, with *Syncarpia glomulifera, Corymbia intermedia, Lophostemon confertus, Allocasuarina torulosa* and *Banksia integrifolia* emergents and co-dominants, of the wet to moist uplands and highlands on yellow earths derived from granitic parent material.	Not of Concern	Not of Concern
7.12.16	8	Simple notophyll vine forest on cloudy wet granite and rhyolite uplands and highlands.	Not of Concern	Not of Concern
7.12.19	9	Simple microphyll vine forest on cloudy wet granite highlands.	Not of Concern	Not of Concern
7.12.21	**14a**	**Tall open rose gum (*Eucalyptus grandis*) forest on cloudy moist granite and rhyolite uplands and highlands.**	**Of Concern**	**Of Concern**
7.12.22	**14b**	**Tall open red mahogany (*Eucalyptus resinifera*) on moist granite and rhyolite uplands and highlands**	**Of Concern**	**Of Concern**

Table 2.4. Change in site health ratings in the period 2000-2002.

Site Code	Severity of Dieback		Tree Fall Damage		Site Drainage		Disturbance of Soil		% Samples at site with positive *P. cinnamomi* response.	
	2000	2002	2000	2002	2000	2002	2000	2002	2000	2002
PO01	2	2	3	1	2	2	0	0	88.89	0
PO02*	N/A	3	N/A	2	N/A	2	N/A	0	N/A	0
PO03*	N/A	3	N/A	4	N/A	2	N/A	0	N/A	62.5
PO04*	N/A	2	N/A	2	N/A	2	N/A	0	N/A	62.5
JP04* (Pilot Study site)	N/A	3	N/A	3	N/A	2	N/A	0	N/A	N/A
PO05	1	2	3	2	2	2	0	0	34.57	37.5
PO06	1	2	2	1	2	2	0	0	97.53	12.5
PO07	2	N/A	3	N/A	1	N/A	1	N/A	67.90	N/A
CO01	0	0	2	1	2	2	0	0	59.26	0
CO02	1	0	3	1	2	2	0	0	41.97	12
CO03*	N/A	0	N/A	2	N/A	1	N/A	0	N/A	25
CO04	0	0	2	2	1	1	1	0	60.49	0
CO05	0	0	2	2	2	2	1	0	56.79	25
CO06	0	0	2	2	2	2	0	0	61.73	50
CO07	0	N/A	2	N/A	2	N/A	0	N/A	55.55	N/A

* Indicates sites whose locality was changed between 2000 and 2002, or that were completely new sites in 2002. See Table 2.2 for more details

Soils

Soil descriptions for sites in the Tully Falls/Koombooloomba area are summarised in Table 2.5. The topsoils (A1 horizon) at all sites are typical of rainforest soils in being acidic, with no differences between affected and unaffected areas.

An association of soil field texture (which is related to clay content) and site was noted. Hypothesis 1, that affected sites show a greater level of change towards heavy clay than unaffected sites, would appear to be supported at these study sites. While the surface (A1, A2, A3) horizons generally had field texture consistent with a sandy clay loam, the clay content became progressively higher with depth in all affected sites except PO01, varying by as much as five grades (Table 2.5). In contrast, the variation observed in unaffected sites was consistently much lower.

Table 2.5. Field texture of soil horizons from profiles dug at study sites in the Koombooloomba Area. Ten of the fifteen texture grades of (McDonald and Isbell, 2000)are shown here. Shading provides an indication of soil depth, darker shading indicating increasing depth in the profile.

Field Texture (in order of increasing clay content).	Affected Sites						Unaffected Sites					
	PO01	PO02	PO03	PO04	PO05	P006	CO01	CO02	CO03	CO04	CO05	CO06
pH	4.7	5.4		4.7	4.9	4.9	4.7	4.7	5.2	4.8	4.7	4.9
Clayey sand				A1								
Sandy loam				A3	A							
Loam												
Silty loam							A1, A3					
Sandy clay loam	A1, B1, B2		A1	B1	B1	A1				A1, A3, B1	A1, A3	A3, C
Clay loam		A1					B11, B21, B22					
Clay loam, sandy			A2, B1			A3, B1						A1
Silty clay loam									A1, A3, B1			
Light clay		B2						A1	B2			
Light medium clay		B1										
Change in Field Texture Grade	0	4	2	6	3	2	2	0	1	1	1	2

2.3.2 SITE DESCRIPTIONS, LAMB RANGE

Detailed descriptions for sites in the Lamb Range area are presented in Table 2.6. Study sites were located along creek flats in the upper catchment of Kauri Creek, within 500m of the Kauri Creek Road (Figure 2.5). Despite careful navigation and extensive searching along mapped and unmapped logging tracks, severe dieback could not be found in other areas of the Lamb Range.

Landform

The Lamb Range is an extensive upland-highland area of rolling to steep hills situated on the Tinaroo Granite (Bain and Draper 1997). The study sites were located at the foot of steep sided, gullies. Creek flats were up to 50 m wide, occasionally somewhat swampy with shallow water tables. All sites were located between 1000 and 1100 m altitude. More detailed descriptions are given in Table 2.6.

Forest Structure

The forest in the upper Kauri Creek catchment has been mapped as Type 8/9 by Tracey and Webb (1975) – a mixture of simple notophyll vine forest and microphyll vine forest. In the deep sheltered gullies where the study sites were located, the forest type is Type 8 dominated, although *Agathis atropurpurea*, characteristic of this forest type was rarely sighted. Sattler and Williams (1999) classify this as RE 7.12.16 (Table 2.7).

The mapped dieback polygons of Stanton and Stanton fall within a variety of forest types – those in the Lamb Range area are listed in Table 2.7. Those Res of conservation concern are highlighted.

Table 2.6. Physical description of sites in the upper catchment of Kauri Creek, in the Lamb Range.

Site Code & Locality Name	Survey Date	Grid Co-ordinates	Altitude	Landform	Forest Type
LRU1 Lamb Range, upper catchment of Kauri Creek.	19-May-02	349275, 8019050	1040m	Creek flats along base of deep gully. Locally very low relief: undulating rises on a gently inclined slope. Landscape consists of high rolling hills with moderately inclined to steep slopes. Southerly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Some evidence of logging activities present, but inconspicuous. Ground ferns, climbing ferns, slender lianes and shrubs prominent in understorey. Canopy height to 40m.				
LRA1 Lamb Range, upper catchment of Kauri Creek	18-May-02	348975, 8108700	1020m	Creek flats along base of deep gully. Locally very low relief: undulating rises on a gently inclined slope. Landscape consists of high rolling hills with moderately inclined to steep slopes. Easterly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Evidence of disease abundant and conspicuous. Little evidence of logging in immediate vicinity. Ground ferns and shrubs prominent in understorey. Weeds (*Rubus alceifolius*) present. Canopy height to 30m.				
LRA2 Lamb Range, upper catchment of Kauri Creek	19-May-02	349152, 8108902	1020m	Creek flats along at junction of two streams. Locally very low relief: undulating rises on a gently inclined slope. Landscape consists of high rolling hills with moderately inclined to steep slopes. Southerly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Abundant evidence of logging, including snig tracks and culvert across creek. Evidence of severe disease. Slender lianes, ground ferns and epiphytes prominent. Canopy height to 25m. Prominent canopy species: *Alphitonia petriei, Polyscias murrayi.*				

Site Health

Both affected sites had unhealthy plants in the canopy and in the understorey, and could be easily related to the mapped dieback polygons of Stanton and Stanton. As at some Mount Lewis sites, the soil disturbance recorded for LRA2 (Table 2.8) was a result of the site being located across a disused snig track. The poor site drainage at this site may have been influenced by construction of logging infrastructure, however, such infrastructure was not present at the other two sites, which also had poor to moderate site drainage.
P. cinnamomi was isolated from soils at all study sites (Table 2.8).

Soils

Because of their proximity to the creek, water was struck within 75 cm of the surface at all sites, and blue-coloured clays, not seen in any other area, were struck in the lowest horizon at both affected sites. The lower profiles of soils at these three sites would therefore appear to be consistently waterlogged. Hypothesis 1 does not appear to be supported by data from study sites in the Lamb Range area, however these sites are unique in this project because of their position on waterlogged creek flats, compared to the ridge and hillslope locations of

other study sites. Comparisons of soil structure with other sites are therefore unlikely to be useful.

Sites LRA2 and LRU1 showed a tendency for increasing clay content through the soil profile, however this was not present at LRA1 (Table 2.9).At this site, a decrease in soil clay with depth was observed, hence a negative value is returned for this site.

Table 2.7. Regional Ecosystems containing mapped dieback polygons in the Lamb Range area.

Regional Ecosystem (Sattler & Williams 1999)	Tracey & Webb (1975) Forest Type	Description (Sattler and Williams 1999)	Biodiversity Status	Conservation Status (VMR)
7.8.1	**1a**	Complex mesophyll vine forest on very wet and wet lowlands and foothills, on krasnozem soils derived from basalts and basic volcanic parent material.	**Of Concern**	**Of Concern**
7.8.2	**1b**	Complex mesophyll vine forest of the very wet and wet cloudy uplands on basaltic krasnozem and euchrozems.	**Endangered**	**Endangered**
7.12.1	2a	Mesophyll vine forest on very wet to wet granite lowlands and foothills.	Not of Concern	Not of Concern
7.12.7	6	Complex notophyll vine forests with emergent *Agathis robusta*, of the moist foothills and uplands on granites.	Not of Concern	Not of Concern
7.12.14	13c	Notophyll vine forest with rose gum (*Eucalyptus grandis*) emergents on cloudy wet granite and rhyolite upland ridges.	Not of Concern	Not of Concern
7.12.16	8	Simple notophyll vine forest on cloudy wet granite and rhyolite uplands and highlands.	Not of Concern	Not of Concern
7.12.19	9	Simple microphyll vine forest on cloudy wet granite highlands.	Not of Concern	Not of Concern

Table 2.8. Site health ratings for Lamb Range study sites.

Site Code	Severity of Dieback	Tree Fall Damage	Site Drainage	Disturbance of Soil	% Samples at site with positive *P. cinnamomi* response.
LRU1	0	1	2	1	12.5%
LRA1	3	2	1	2	25%
LRA2	2	1	1	2 (snig track)	12.5%

Table 2.9. Field texture of soil horizons from profiles dug at study sites in the Lamb Range area. Twelve of the fifteen texture grades of (McDonald and Isbell, 2000)are shown here. Shading provides an indication of soil depth, darker shading indicating increasing depth in the profile.

Field Texture (in order of increasing clay content).	Unaffected Sites	Affected Sites	
	LRU1	LRA1	LRA2
Sand		A2	A1
Loamy sand			
Clayey sand	A1, A2	B1	
Sandy loam			
Loam		A1	
Silty loam			B1
Sandy clay loam	?B2		
Clay loam			
Clay loam, sandy			
Silty clay loam			C1
Light clay			
Light medium clay	B1		
Change in Field Texture	9	-4	9

2.3.3 SITE DESCRIPTIONS, MOUNT LEWIS

In contrast to the Tully Falls/Koombooloomba area, no study sites had been established at Mt Lewis, therefore site selection aimed to find visible dieback patches within mapped dieback polygons, and use these as study sites. Site selection is further discussed in Section 1.3.2. The tasks carried out at each site are detailed in Table 2.1

Site MLU4 served as a control (unaffected site) for sites MLA4, MLA4b and MLA5.

Landform

Mt Lewis lies at the southern end of the Carbine Tableland, a granite massif with summits rising to >1000 m. Slopes in the study area range from gentle to very steep (Table 2.10) The majority of study sites were located on ridgelines and adjoining slopes. MLA3 and MLA2 were located in gullies.

Forest Structure

Most of the mapped dieback polygons in the Mt Lewis area fall within Type 8/Type 9 forest, however, the forest around MLA14 was assessed as Type 2a forest. These forest types are described in Table 2.11.

Site Health

All sites were located within 500m of the Mount Lewis Road, and often were located on snig tracks. Soil disturbance associated with snig tracks was noted (Table 2.12).

Dieback severity at affected sites generally moderate, except for site MLA4b, which displayed severe dieback. Red-brown resinous cankers were observed on unhealthy trees at this site (Figure 2.7). *P. cinnamomi* was isolated from most sites, although it was not isolated in two apparently affected sites (MLA3 and MLA5).

Site MLA6 was located adjacent to the CSIRO study plot, within a recovering dieback site. Although the canopy at this site was open, a dense growth of understorey saplings suggested the site was recovering.

Table 2.10. Physical description of sites in the Mount Lewis area.

Site Code & Locality Name	Survey Date	Grid Co-ordinates	Altitude	Landform	Forest Type
MLU1	23-Oct-01	318307, 8165290	867m	Not recorded.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Located on an old snig track. Canopy height to 35m. Ferns prominent in understorey. Canopy height to 30m.				
MLU4	Feb-02	316800, 8163820	960m	Not recorded.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Located uphill from MLA4, on old snig track. Canopy to 30m high.				
MLU6 (adjacent to CSIRO permanent plot)	9-Nov-02	314800, 8172265	1080m	Hillslope on undulating low hills. North westerly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Prominent canopy species: *Syzygium wesa, Elaeocarpus largiflorens ssp. largiflorens, Beilschmiedia bancroftii & Beilschmiedia collina.*				
MLA1	12-Jan-02	318307, 8165290	875 m	Moderately inclined to steep hillslope and hillcrest on rolling high hills. West-south-westerly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Located on an old snig track, only a short distance from the Mt Lewis Road.. Canopy height to 35m. Ferns prominent in understorey. Prominent canopy species: *Pouteria euphlebia, Syzygium kuranda & Balanops australiana.*				
MLA2	23-Oct-01	317250, 8165300	870m	Hillcrest on rolling to steep low hills. North-north easterly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	No evidence of anthropogenic disturbance. *Oraniopsis* palms prominent in the understorey. Canopy height to 40m.				
MLA4	24-Oct-01	316963, 8164251	980m	Hillcrest on rolling hills. Southerly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Site contains tracks and evidence of logging. Canopy to 20m high.				
MLA5	1-Nov-01	315050, 8164150	940m	Moderately inclined hillslope on high rolling hills. South-westerly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Site lies near end of abandoned snig track. Severe dieback. Canopy to 20m. Prominent canopy species: *Flindersia bourjotiana, Sloanea macbrydei, Cardwellia sublimis.*				
MLA6 (adjacent to CSIRO permanent plot)	Feb-02	314800, 8172265	1080m	Hillslope on undulating low hills. Southerly aspect..	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	One of the first dieback sites reported in the Wet Tropics. Vegetation consists mostly of regrowth on an old dieback site. Canopy to 30m.				
MLA14 Mt Lewis	13-Feb-02	315812, 8168582	1040m	Gently inclined hillslope on undulating low hills. Westerly aspect.	Type 2a (Mesophyll Vine Forest) RE 7.12.1
	No evidence of disease recorded within actual site, however, roadbuilding appears to have altered drainage patterns at a nearby creek flat, causing swampiness. Several trees in this area were sick or dead. Site located partly on an abandoned logging camp/log loading area. Regrowth dominated by Cunoniaceae. Prominent canopy species: *Elaeocarpus eliffii, Sloanea macbrydei & Syzygium gustavioides.*				

Table 2.11. Regional Ecosystems containing mapped dieback polygons in the Mount Lewis area.

Regional Ecosystem	Tracey & Webb (1975) Forest Type	Description (Sattler and Williams 1999)	Biodiversity Status	Conservation Status (VMA)
7.12.1	2a	Mesophyll vine forest on very wet to wet granite lowlands and foothills.	Not of Concern	Not of Concern
7.12.16	8	Simple notophyll vine forest on cloudy wet granite and rhyolite uplands and highlands.	Not of Concern	Not of Concern
7.12.19	9	Simple microphyll vine forest on cloudy wet granite highlands.	Not of Concern	Not of Concern

Table 2.12. Site health ratings for Mount Lewis study sites.

Site Code	Severity of Dieback	Tree Fall Damage	Site Drainage	Disturbance of Soil	% Samples at site with positive *P. cinnamomi* response.
MLU1	0	1	3	2 (snig track)	12.5%
MLU2	0	1	3	0	25%
MLU3	0	1	2	0	0%
MLU4	0	1	3	2 (snig track)	0%
MLU6	0	1	3	0	18.8%
MLA1	2	2	3	2 (snig track)	50%
MLA2	1	2	3	0	18.8%
MLA3	2	3	1	0	0%
MLA4	2	1	3	2 (snig track)	18.8%
MLA4b	3	2	3	0	50%
MLA5	2	1	3	2 (snig track)	0%
MLA6	3 (recovering)	4	2	0	18.8%
MLA14	0	1	2	2 (snig track)	-
MLU1	0	1	3	2 (snig track)	12.5%
MLA4b	3	2	3	0	50%
MLA5	2	1	3	2 (snig track)	0%
MLA6	3 (recovering)	4	2	0	18.8%
MLA14	0	1	2	2 (snig track)	-

Soils

Soils at affected sites in the Mt Lewis area showed some increase in soil clay through the profile, however this increase does not appear to be significant when compared with the profiles from unaffected sites (Table 2.13). Hypothesis 1 does not appear to be supported by data from the Mt Lewis study sites.

Table 2.13. Field texture of soil horizons from profiles dug at study sites in the Mount Lewis Area. Ten of the fifteen texture grades of (McDonald and Isbell, 2000) are shown here. Shading provides an indication of soil depth, darker shading indicating increasing depth in the profile.

Field Texture (in order of increasing clay content).	Affected Sites					Unaffected Sites		
	MLA1	MLA2	MLA3	MLA4	MLA14	MLU1	MLU3	MLU4
Phytophthora	4.7	4.6	5.1	5.3	4.9	4.7	4.5	4.8
Clayey sand								
Sandy loam								
Loam					A1			
Silty loam								
Sandy clay loam	A1	A1	A1	A1, A2, A3, B2	A2	A1		
Clay loam							A1	
Clay loam, sandy	A3, B2, B3, C1	A3, B2, B3, C	A2, A3		B2	A2, A3, B1	A2, A3	A1, A3, B1, B2
Silty clay loam								
Light clay						B2		
Light medium clay								
Change in Field Texture	2	2	2	0	4	4	1	0

2.3.4 SITE DESCRIPTIONS, KIRRAMA

Mapped dieback polygons in the Cardwell Range, near Kirrama, proved extremely difficult to locate. Sites were eventually identified along or adjacent to a snig track which traversed a west-trending ridge leading from the abandoned logging camp at Gunna Camp. The approximate location of Gunna Camp is shown in Figure 2.6.

Figure 2.14. Physical description of sites in the Cardwell Range, near Kirrama.

Site Code & Locality Name	Survey Date	Grid Co-ordinates	Altitude	Landform	Forest Type
KIU1 Gunna Camp, near Kirrama	4-Oct-02	372160, 7982995	920m	Moderately inclined hillslope on rolling low hills. North easterly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Site located on a disused snig track. Evidence of logging occasional but not conspicuous. Dead canopy trees present, but no dead or dying trees in understorey. Climbing pandans, tree ferns and ground ferns abundant in understorey. Canopy to 35m. Prominent canopy species include: *Flindersia bourjotiana*, *Sloanea macbrydei.*				
KIA1	2-Oct-02	372408, 7983098	920m	Moderately inclined hillslope on rolling low hills. South-south easterly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Site located on a disused snig track. Evidence of logging occasional but conspicuous. Slender lianes abundant in the understorey. Canopy to 30m.				
KIA2 Gunna Camp, near Kirrama	4-Oct-02	372034, 7982654	960m	Moderately inclined hillslope on rolling low hills. South-south westerly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Site located on a disused snig track. Evidence of logging occasional but conspicuous. Shrubs, ground ferns and climbing pandans abundant in the understorey. Canopy to 40m. Prominent canopy species: *Franciscodendron laurifolium.*				

Landform

Land forms in the area consist of rolling to steep hills, situated on granites of the Ingham Batholith (Bain and Draper 1997). Drainage lines tend in a southerly direction.

Forest Structure

As with other study sites, the mapped dieback polygons were located in a variety of forest types (Table 2.15). All study sites were located in Type 8 forest.

Table 2.15. Regional Ecosystems containing mapped dieback polygons in the Kirrama/Cardwell Range area.

Regional Ecosystem	Tracey & Webb (1975) Forest Type	Description (Sattler and Williams 1999)	Biodiversity Status	Conservation Status (VMA)
7.8.1	**1a**	Complex mesophyll vine forest on very wet and wet lowlands and foothills, on krasnozem soils derived from basalts and basic volcanic parent material.	**Of Concern**	**Of Concern**
7.12.1	2a	Mesophyll vine forest on very wet to wet granite lowlands and foothills.	Not of Concern	Not of Concern
7.12.14	13c	Notophyll vine forest with rose gum (*Eucalyptus grandis*) emergents on cloudy wet granite and rhyolite upland ridges.	Not of Concern	Not of Concern
7.12.16	8	Simple notophyll vine forest on cloudy wet granite and rhyolite uplands and highlands.	Not of Concern	Not of Concern
7.12.19	9	Simple microphyll vine forest on cloudy wet granite highlands.	Not of Concern	Not of Concern

Site Health

The snig track heading west from Gunna Camp was inspected for a distance of approximately 2.5km. Dieback was not severe in any of the forest traversed (Table 2.16). Several dead canopy trees were observed at site KIA1, but there was little evidence of disease in the understorey. Site KIA2, judged to be the worst affected point within a mapped

dieback polygon, similarly had dead canopy trees and a nearby canopy gap but a healthy understorey.

P. cinnamomi was isolated from affected sites, but not from the unaffected site.

Table 2.16. Site health ratings for Kirrama study sites

Site Code	Severity of Dieback	Tree Fall Damage	Site Drainage	Disturbance of Soil	% Samples at site with positive *P. cinnamomi* response.
KIU1	0	1	3	2 (snig track)	0
KIA1	2	1	3	2 (snig track)	12.5%
KIA2	1	2	3	0	12.5%

Soils

Profile descriptions are given in Table 2.17. As with other sites, there was a tendency toward greater levels of clay with increasing depth. There was not, however, clear differences between affected and unaffected sites. Hypothesis 1 does not appear to be supported by data from Kirrama study sites.

Table 2.17. Field texture of soil horizons from profiles dug at study sites in the Kirrama area. Ten of the fifteen texture grades of (McDonald and Isbell, 2000)are shown here. Cell shading is indicative of soil depth, darker shading indicating increasing depth in the profile.

Field Texture (in order of increa-sing clay content).	KIU1					KIA1					KIA2			
	Profile 1	Profile 2	Profile 3	Profile 4	Profile 5	Profile 1	Profile 2	Profile 3	Profile 4	Profile 5	Profile 1	Profile 2	Profile 4	Profile 5
Topsoil pH	5.3	5.6	5.6	5.4	5.1	5.7	5.3	5.4	5.4	5.6	5.5	5.6	5.4	5.1
Clayey sand														
Sandy loam			A	A	A			A	A	A				
Loam														
Silty loam														
Sandy clay loam	A, B1, B21	A, B1, B2	B1, B2, B3, C	B1, B21 B22 B3	B1, B2, B3	A, B1, B21, B22	A, B1, B21, B22	B1, B2, B3	B1, B2, B3	B1, B21	A, B1, B2	A, B1	A, B1	A, B1
Clay loam														
Clay loam, sandy	?B22 ?B3	?B3			?C		?B3			B22, B3	?B3, C	?B2 ?B3	?B21 B22	?B2, ?B3
Silty clay loam														
Light clay														
Change in Field Texture	2	2	3	3	5	0	2	3	3	5	2	2	2	2

2.3.5 SITE DESCRIPTIONS, OTHER AREAS

In addition to the sites mapped by Stanton and Stanton (Figure 2.1), reports have been received of dieback-like symptoms in several localities, mostly in upland areas of the Wet Tropics. Some of these have been visited, and are described briefly in Table 2.18.
Other uninvestigated reports of dieback-like patch death have come from:

- Francis Range, near Bartle Frere
- in eucalypt woodland near Mount Fox, west of Rollingstone
- above the overflow of Copperlode Dam, Cairns
- on basalt soils near Topaz

Table 2.18. Physical description of other patch-death sites observed during 2001-2002

Site Code & Locality Name	Survey Date	Grid Co-ordinates	Altitude	Landform	Forest Type
MMA1 Mt Mackay Carbine Tableland	23-Jun-02	316083, 8160223	1165m	Gently sloping ridge on rolling high hills.	Type 8 (Simple Notophyll Vine Forest)/ Type 9 (Simple Microphyll Vine-Fern Forest) RE 7.12.16 / 7.12.19
	The largest dieback patch observed during this project (see frontispiece). Active, severe dieback (Dieback severity rating = 3). Soils derived from granite (boulders present on edge of patch). This site was mapped by Stanton and Stanton. (Figure 2.10)				
Shannessy Creek, Mt Spurgeon, Carbine Tableland	14-Nov-02	~307800, 8181700	~1200m	Gently sloping ridge on rolling high hills.	Type 14a (Tall open *Eucalyptus grandis* forest.) RE 7.12.21
	Soils sampled and baited. No *Phytophthora* isolated. Dead and dying *Eucalyptus grandis* in ecotone forest north-west of Mt Spurgeon. No other species suffering symptoms of dieback. No progression of disease down hills. No recently dead leaves hanging in canopy. *Phytophthora cinnamomi* ruled out as cause of dieback at this si. Other contributing factors/causes may be insects (borers and/or folivores), long-term drought, *Armillaria* fungus, changes in fire regime.				
Black Mtn (Harris Peak)	Dec-01	~339500, 8158000	~900 m	Gently undulating ridgeline with steep slopes either side. Numerous granite boulder outcrops along ridgeline. South-easterly aspect.	Type 8 (Simple Notophyll Vine Forest) RE 7.12.16
	Soils not sampled. Two small patches on ridge track at about 800-900 m. This site had not previously been reported. Dieback moderate to severe (Dieback severity rating = 3), with some trees showing signs of canopy thinning.				
"Devil's Thumb" access track, Karnak[1]	Sep-02	~318000, 8186600	~1050 m	Gently undulating ridgeline. Numerous granite boulder outcrops along ridgeline.	Type 10 (Simple Microphyll Vine-Fern Thicket) RE 7.12.20
	Several small patches along ridge track at about 1000-1100 m. Dieback patches contain recently dead trees and appear to be expanding (Dieback severity rating = 3). Soils sampled. Isolates not yet identified.				
Mount Bartle Frere, western access track[2]	Dec-02	~373600, 8076750	~1520 m	Undulating ridgeline. Numerous granite boulder outcrops along ridgeline.	Type 9 (Simple Miicrophyll Vine-Fern Forest) Type 10 (Simple Microphyll Vine-Fern Thicket) RE 7.12.19/7.12.20
	Soils not sampled. Several distinct dieback patches (Dieback severity rating = 3) along perhaps a kilometre of the summit track, between the North-West Peak and the South Peak. The grid reference is the southernmost patch. (Figure 2.9)				

[1] Devil's Thumb an unofficial name for a rocky pinnacle on the Main Range, west of Karnak. The official Devil's Thumb is located approximately 7 km south of the grid reference listed here.
[2] During a visit to Mt Bartle Frere in January 2003, individuals of *Eucryphia wilkiei* (listed as Vulnerable under the Queensland *Nature Conservation Act 1994*) on the main summit access track were observed to be suffering symptoms similar to those caused by *P. cinnamomi* infection.

Dieback in Eucalyptus grandis at Mount Spurgeon

In the Mt Spurgeon area, *Eucalyptus grandis* is suffering from dieback, with many trees declining in vigour, dying or dead. Reports of a similar problem have been received from the Princess Hill area of Lumholtz National Park, west of Ingham (Paul Williams personal communication). *E. grandis* in the Spurgeon area has never been logged, although some disturbance associated with mining may have occurred.

Observed symptoms and characteristics of the problem include:

- decline in vigour
- loss of canopy
- some evidence of shooting from epicormic buds along main branches (but not along trunk)
- no dead leaves hanging in canopy – therefore no evidence of recent active dieback
- dead/dying trees mostly mature (> 60 cm dbh), although some smaller trees suffering
- no affected saplings observed
- dead/dying trees appear to be randomly distributed through the landscape (although there were possibly more dead trees lower in the landscape)
- several dead/dying trees were observed within 30m of a flowing creek (therefore, drought is unlikely to be playing a role)

- there was no evidence of a "disease front" – a wave of disease passing through the landscape, leaving dead trees in its wake, and slowly killing trees at the front
- no other species appeared to be affected (see attached species list).

Possible causes for the dieback include:

1. *Phytophthora cinnamomi*
Unlikely, as all other species in the community were healthy. Also, dead/dying trees appeared randomly dispersed through the community, rather than following a "disease front" pattern of infection that characterises *Phytophthora cinnamomi*-related dieback.
Cardwellia sublimis, which is listed as a susceptible species (Table 2.22), was observed healthy and flowering downhill from affected *E. grandis*. As *P. cinnamomi* is dispersed by water, one might expect susceptible species downhill from affected plants to also be affected.

2. Long term drought
Unlikely, as some dead/dying trees were situated quite low in the landscape, close to drainage lines and flowing streams. A difficult hypothesis to test.

3. *Armillaria* or some other wood-rotting fungus
There was no evidence of active or recently active dieback in the affected trees (i.e. there were no dead or recently dead leaves hanging in the trees). If a species of wood-rotting fungus is responsible for the dieback, the recent drought may have slowed the activity of the fungus in affected trees.

4. Changes in fire regime

5. Insect attack

Landform
The sites described in Table 2.18 are located on high altitude ridgelines, frequently with thin soils and boulder outcrops. Except for the Mt Mackay site, slopes to either side of the ridge alignments are steep to very steep. All are situated on granites. The ridgelines described carry walking tracks, although two of these (Black Mountain and Mount Mackay) are infrequently used. The Mount Spurgeon is unlikely to be utilised as a walking track.

Forest Structure
Except for the tall open forest at Mount Spurgeon, the forest structure is typical of that occurring on granitic highlands in the Wet Tropics, with canopy heights from 8 to 25 m, frequent outcrops of granite boulders and thin soils. Forest types observed at the sites in Table 2.18 are described below (Table 2.19).

Site Health
All sites could be described as suffering severe to very severe dieback (Dieback severity rating = 3). Details of soil disturbance and treefall damage were not described. As these sites are located on ridges, it is likely they will be freely draining.

Soils
Not described.

Table 2.19. Regional Ecosystems containing patch death sites described in Table 2.18.

Regional Ecosystem	Tracey & Webb (1975) Forest Type	Description (Sattler and Williams 1999)	Biodiversity Status	Conservation Status
7.12.16	8	Simple notophyll vine forest on cloudy wet granite and rhyolite uplands and highlands.	Not of Concern	Not of Concern
7.12.19	9	Simple microphyll vine forest on cloudy wet granite highlands.	Not of Concern	Not of Concern
7.12.20	**10**	**Low microphyll rainforest on cloudy moist granite and rhyolite uplands and highlands.**	**Of Concern**	**Of Concern**
7.12.21	**14a**	**Tall open rose gum forest on cloudy moist granite and rhyolite uplands and highlands.**	**Of Concern**	**Of Concern**

2.3.6 CANOPY ASSESSMENTS

Hemispherical Photography – Pilot Study

Hemiphots were taken at 73 photopoints in the pilot study site (JP04) near Tully Falls. Of these, 41 proved suitable for analysis. Others were rejected for one or more of the following reasons:

- photos were overexposed
- photos were underexposed
- sunlight shining directly on understorey vegetation caused bright patches, which the Winphot program interprets as canopy gaps and holes (eg. Figure 2.11c) This lead to an unrealistically high value of the canopy openness

Examples of hemiphots, from the central point of each of the Tully Falls/Koombooloomba sites, are given in Figure 2.11a-f. The mean canopy openness for each study site is shown in Figure 2.12.

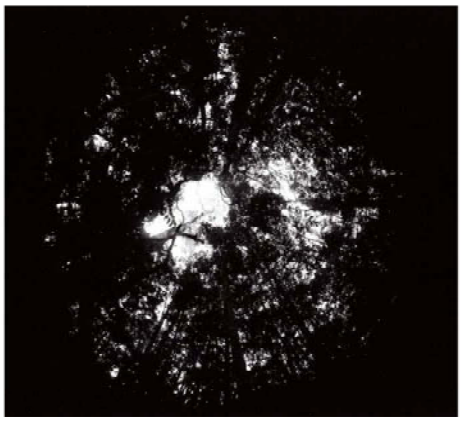
Hemispherical Photography - Analyses

The mean canopy openness for each study site is shown in Figure 2.12. Comparisons of affected and unaffected sites indicated there were no differences in CO between affected and unaffected sites (ANOVA: $F_{1,12} = 1.37$, $P = 0.27$). Hypothesis 1 is not supported.

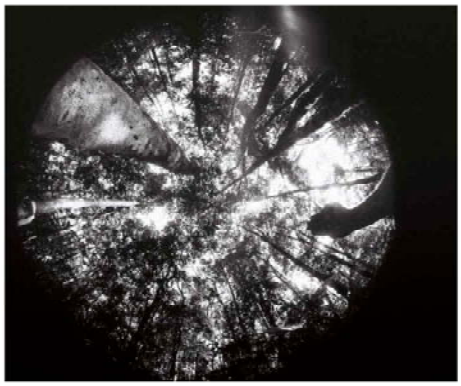
Some trends can be observed in the data. The differences between affected and unaffected sites in the Lamb Range area are marked Figure 2.12). In the Tully Falls/Koombooloomba area, the Pilot Study site and PO03 had the most visibly severe and widespread symptoms of dieback, and were allocated the highest dieback severity ratings (Table 2.4). Despite being located within mapped dieback polygons, PO05 and PO06 did not have higher canopy openness, and their dieback severity ratings reflect this.

Gaps proved difficult to interpret using this methodology. Although canopy thinning was evident in hemiphots, actual canopy gaps were rare. Increases in canopy openness observed were due to canopy thinning, not canopy gaps. The number and size of canopy gaps at study sites could not be quantified, therefore hypotheses 3 and 4 could not be addressed using this method.

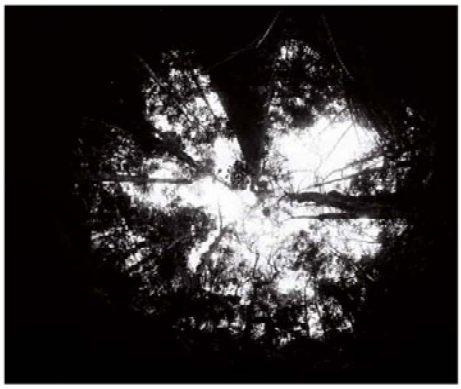
a) CO03. Openness = 15.18%



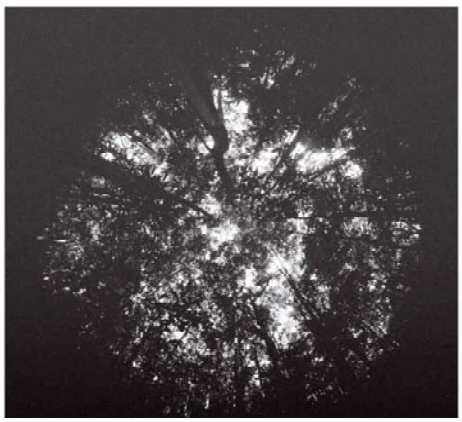
b) CO04. Openness = 19.74%



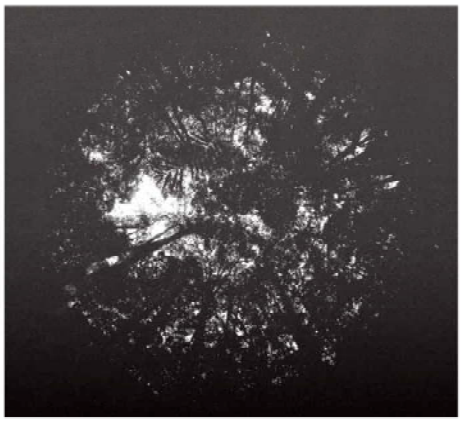
c) Pilot Study (Site JP04) Understory light levels too high, and sky too glary - canopy openness could not be calculated for this site.



d) PO03. Openness = 25.88%



e) PO05. Openness = 20.42%



f) PO06. Openness = 15.50%

Figure 2.11 a-f. Canopy hemiphots taken at the central photopoint of each study site in the Koombooloomba/Tully Falls area. Site code and canopy openness are indicated.

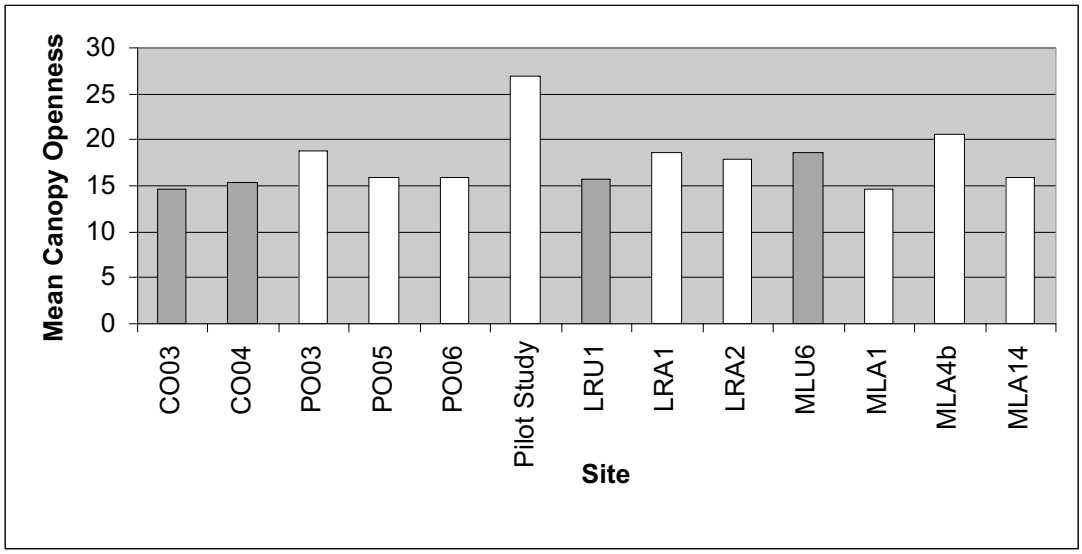


Figure 2.12. Mean canopy openness derived from hemiphots. Shaded bars indicate unaffected sites.

2.3.7 FLORISTICS

Across all study sites, a total of 3565 canopy, subcanopy and understorey trees were assessed. Nine hundred and thirty five of these stems were located on unaffected sites. A total of 248 species in 52 families were assessed. A further 68 species (generally non-canopy species) were also recorded at study sites, but not included in analyses. In terms of abundance, by far the most common species was *Flindersia bourjotiana*, with 172 stems counted. The next most common species were *Pullea stutzeri* (109 stems), *Syzygium kuranda* (99 stems), *Fransicodendron laurifolium* (91 stems) and *Cryptocarya mackinnoniana* (85 stems). Forty-nine species were encountered only once. One hundred and thirty eight stems were listed as unknown (*i.e.* species not identified), however, the majority of these were dead and consequently unidentifiable. A full species list area is given in Appendix II.

Stem Densities

There were no significant differences between mean living stem densities between affected and unaffected sites (ANOVA: $F_{1,12} = 0.20$, $P = 0.67$). Nor were there trends for increased or decreased stem densities at affected sites when compared with unaffected sites – stem densities at unaffected sites fell within, or close to, the range of values observed for affected sites (Figure 2.13). Hypothesis 7 (that mapped dieback polygons will have lower living stem densities than unaffected forest) is not supported.

There were no differences between affected and unaffected sites in the mean basal area of living stems (ANOVA: $F_{1,12} = 0.18$, $P = 0.68$), see also Figure 2.14. Hypothesis 5 (that sites within mapped dieback polygons have lower stem basal areas than similar sites in unaffected forest) is not supported. However, when number of stems in each size class were compared, there was a significant difference between affected and unaffected sites ($\chi^2 = 11.864$, df = 3, $P < 0.01$). This difference results from affected sites having a higher proportion of small trees compared to unaffected sites, and affected sites having a higher proportion of smaller size classes (Figure 2.15).

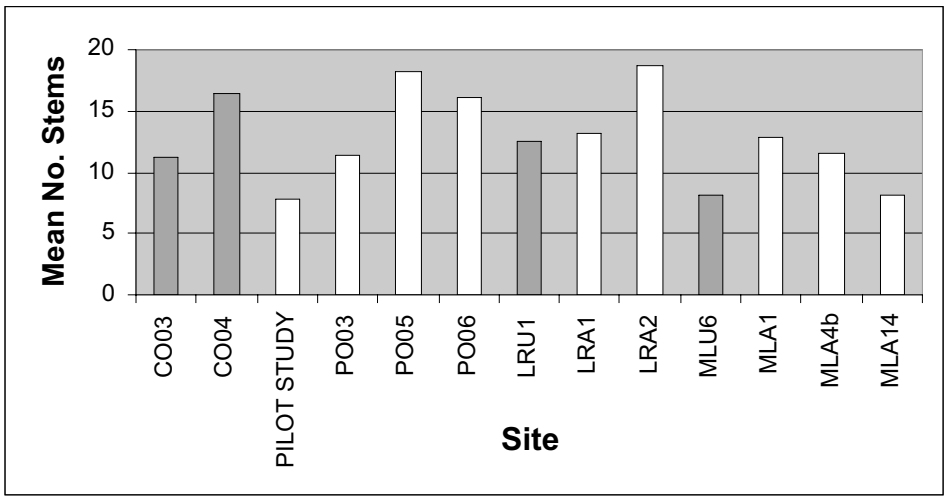


Figure 2.13. Mean number of stems per photopoint at each study site. Unaffected sites are shaded.

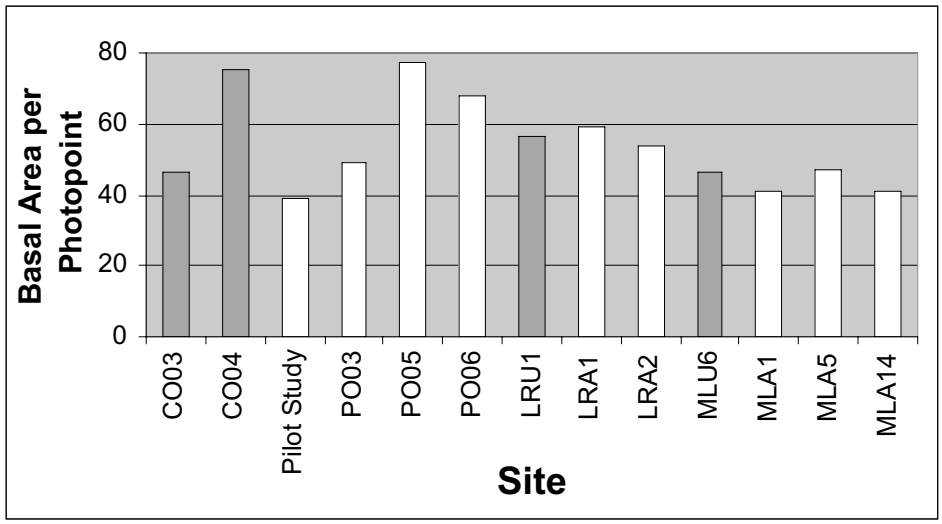


Figure 2.14. Comparison of mean stem basal areas plot at each study site. Stem basal area was calculated from tree size class data, as defined in Section 2.2.5.

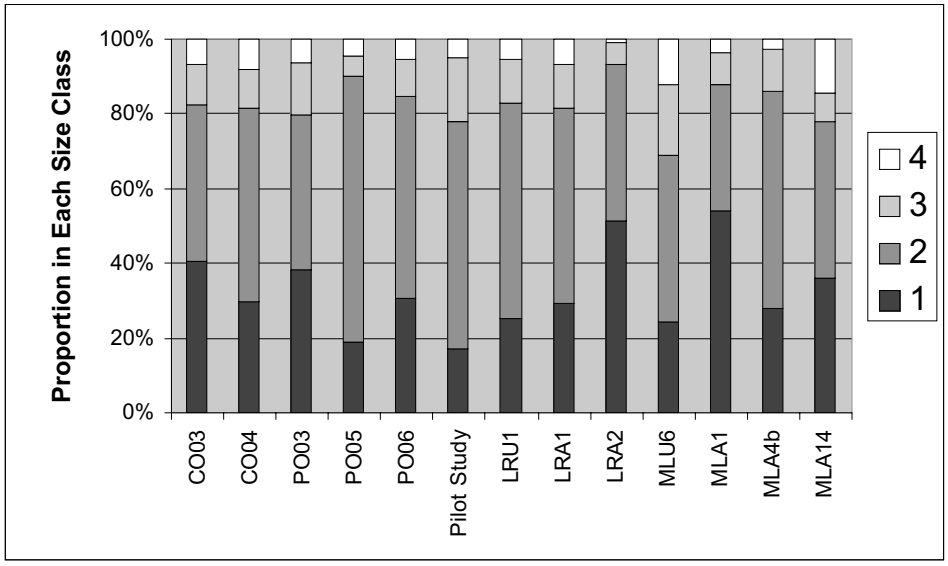


Figure 2.15. Proportion of stems in each size class at each site.

Species Diversity

Diversity for affected sites in all areas did not vary significantly from that of unaffected sites (Figure 2.16). For example, although Site LRU1 has more species, its Shannon-Wiener Index fell between that calculated for the two affected sites. Differences between affected and unaffected sites with respect to diversity were not significant (ANOVA: $F_{1,12} = 0.21$, $P = 0.66$), nor were actual numbers of species (ANOVA: $F_{1,12} = 0.04$, $P = 0.85$).

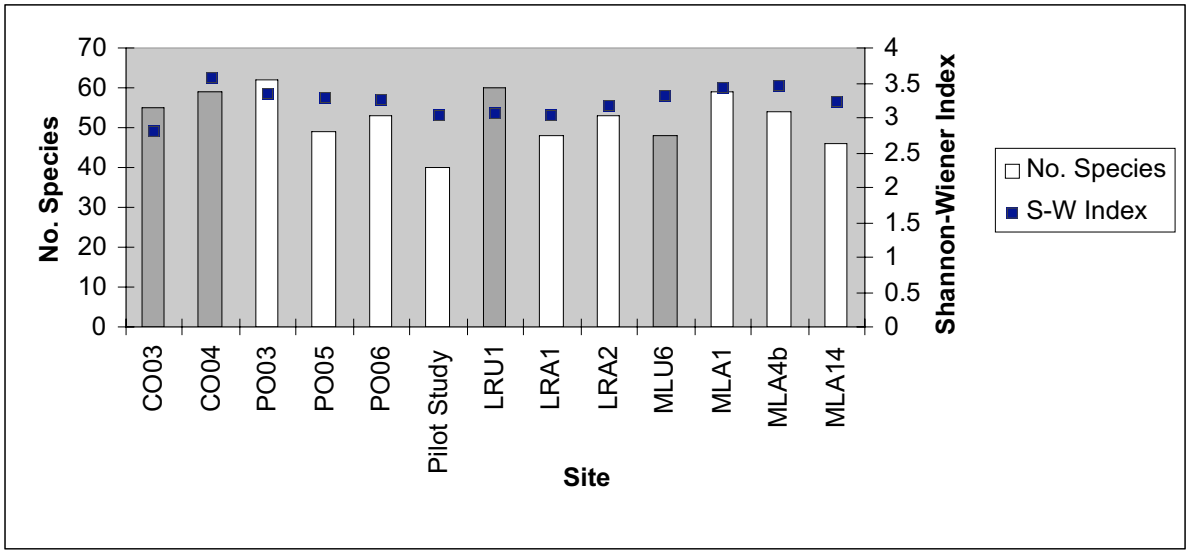


Figure 2.16.Species diversity at each study site, showing the number of species present and the Shannon-Wiener index. Only the number of species in the first sixteen photopoints have been considered in calculations. Unaffected sites are shaded.

Seventy five species were only recorded from affected sites, thirty three were unique to unaffected sites (Table 2.20).st of these species are only represented by one or a few stems – they were rare in our surveys. Table 2.20 utilises all available species records in Appendix II – both stem counts and presence/absence records. Several species previously recorded only from affected sites (Gadek *et al.* 2001) were recorded exclusively from unaffected sites, and vice versa. Amongst common species, there was no indication that selection for resistant or susceptible species was occurring. Thus, species that were common in affected sites were also present in unaffected sites, and *vice versa* (Appendix II).

Successional Status

Figure 2.17 shows the proportion of stems falling into each successional status category. The majority of stems in all sites belonged to species in category 3, (defined as "grows in well-developed rainforest"). Some 318 stems belonged to species that could not be classified (termed "Other" in Figure 2.17). These were excluded from analyses.

Site CO03, in the Tully Falls/Koombooloomba area, had a high proportion of early successional species which are favoured by disturbance, including *Acacia celsa, Acronychia acidula* (this species not found at any other site) and *Darlingia darlingiana*, suggesting recent disturbance. The highest proportion of category 4 species (perhaps indicating undisturbed forest, or forest only recently affected by dieback) were in sites CO04, PO06 and MLA4b. Surprisingly, the Lamb Range unaffected site contained a high proportion of early successional stage species, perhaps resulting from its position in a gully. Evidence of logging and associated soil disturbance, which might be expected to promote a high number of early successional plants, was not evident at this site, although it was prominent at LRA2.
Differences between affected and unaffected sites were significant ($\chi^2 = 55.71$, df = 4, $P < 0.001$). The proportion of stems in early successional class was higher than expected in unaffected sites, as was the proportion of mature forest species. The proportion of class two and three stems was slightly lower in unaffected sites.

Table 2.20. Species which were recorded exclusively from affected, or unaffected sites, and the number of sites from which each species was recorded. Shaded cells indicate species which were also recorded exclusively from affected or unaffected sites by Gadek *et al.* (2001). Cells with double outline were previously recorded exclusively from sites of the opposite disease designation (*e.g.* Gadek *et al.* 2001 reported *Endiandra palmerstonii* from unaffected sites only)

Taxa Recorded in Affected Sites Only			Taxa Recorded in Unaffected Sites only		
Family	**Species**	**No. Sites**	**Family**	**Species**	**No. Sites**
Proteaceae	*Lomatia fraxinifolia*	6	Euphorbiaceae	*Aleurites rockinghamensis*	2
Elaeocarpaceae	*Sloanea macbrydei*	5	Lauraceae	*Cryptocarya grandis*	2
Aquifoliaceae	*Sphenostemon lobosporus*	4	Arecaceae	*Calamus australis*	1
Myrtaceae	*Acmena resa*	4	Burseraceae	*Canarium australasicum*	1
Cunoniaceae	*Gillbeea whypallana*	3	Ebenaceae	*Diospyros cupulosa*	1
Elaeocarpaceae	*Elaeocarpus elliffii*	3	Ebenaceae	*Diospyros pentamera*	1
Lauraceae	*Endiandra sankeyana*	3	Euphorbiaceae	*Drypetes acuminata*	1
Mimosaceae	*Archidendron grandiflorum*	3	Fabaceae	*Castanospermum australe*	1
Proteaceae	*Placospermum coriaceum*	3	Grossulariaceae	*Polyosma rigidiuscula*	1
Rutaceae	*Acronychia acronychioides*	3	Lauraceae	*Cryptocarya cocosoides*	1
Symplocaceae	*Symplocos ampulliformis*	3	Lauraceae	*Cryptocarya saccharata*	1
Elaeocarpaceae	*Elaeocarpus largiflorens ssp. retinervis*	2	Lauraceae	*Endiandra acuminata*	1
Euphorbiaceae	*Macaranga subdentata*	2	Lauraceae	*Endiandra phaeocarpa*	1
Flacourtiaceae	*Casearia costulata*	2	Moraceae	*Ficus crassipes*	1
Lauraceae	*Litsea connorsii*	2	Myrtaceae	*Pilidiostigma tetramerum*	1
Lauraceae	*Endiandra monothyra ssp. trichophylla*	2	Myrtaceae	*Rhodomyrtus macrocarpa*	1
Lauraceae	*Endiandra dielsiana*	2	Myrtaceae	*Syzygium canicortex*	1
Meliaceae	*Synoum muelleri*	2	Proteaceae	*Stenocarpus reticulatus*	1
Meliaceae	*Dysoxylum oppositifolium*	2	Rhamnaceae	*Emmenosperma alphitonioides*	1
Monimiaceae	*Wilkiea wardellii*	2	Rubiaceae	*Canthium costatum*	1
Monimiaceae	*Tetrasynandra laxiflora*	2	Rubiaceae	*Canthium sp. (Herberton Range S.F. Kajewski 1377)*	1
Monimiaceae	*Steganthera macooraia*	2	Rubiaceae	*Cyclophyllum multiflorum*	1
Myrtaceae	*Waterhousea unipunctata*	2	Rubiaceae	*Ixora sp. (North Mary L.A. BH 8618)*	1
Myrtaceae	*Syzygium luehmannii*	2	Rutaceae	*Acronychia acidula*	1
Myrtaceae	*Austromyrtus shepherdii*	2	Rutaceae	*Zanthoxylum veneficum*	1
Pandanaceae	*Pandanus monticola*	2	Sapindaceae	*Arytera pauciflora*	1
Proteaceae	*Musgravea stenostachya*	2	Sapindaceae	*Jagera pseudorhus var. integerrima*	1
Proteaceae	*Carnavonia araliifolia var. montana*	2	Sapindaceae	*Mischocarpus pyriformis ssp. pyriformis*	1
Rhamnaceae	*Schistocarpaea johnsonii*	2	Sapindaceae	*Sarcotoechia protracta*	1
Rubiaceae	*Antirhea tenuiflora*	2	Sapindaceae	*Toechima monticola*	1
Rutaceae	*Acronychia vestita*	2	Sapotaceae	*Pouteria castanosperma*	1
Sapindaceae	*Sarcopteryx montana*	2	Sapotaceae	*Pouteria sp. (Mt Lewis B.P. Hyland 579)*	1
Annonaceae	*Meiogyne sp. (Mt Lewis L.W. Jessup 554)*	1	Urticaceae	*Dendrocnide moroides*	1
Araliaceae	*Polyscias purpurea*	1			
Balanopaceae	*Balanops australiana*	1			
Celastraceae	*Hypsophila halleyana*	1			
Celastraceae	*Hypsophila dielsiana*	1			
Celastraceae	*Hedraianthera porphyropetala*	1			
Clusiaceae	*Garcinia warrenii*	1			
Corynocarpacea	*Corynocarpus cribbianus*	1			

Taxa Recorded in Affected Sites Only			Taxa Recorded in Unaffected Sites only		
Family	**Species**	**No. Sites**	**Family**	**Species**	**No. Sites**
Elaeocarpaceae	*Elaeocarpus bancroftii*	1			
Elaeocarpaceae	*Aceratium ferrugineum*	1			
Euphorbiaceae	*Drypetes iodoformis*	1			
Euphorbiaceae	*Baloghia parviflora*	1			
Flacourtiaceae	*Homalium circumpinnatum*	1			
Gentianaceae	*Fagraea fagraeacea*	1			
Grossulariaceae	*Abrophyllum ornans*	1			
Lauraceae	*Endiandra palmerstonii*	1			
Lauraceae	*Endiandra leptodendron*	1			
Lauraceae	*Endiandra hypotephra*	1			
Lauraceae	*Cryptocarya murrayi*	1			
Lauraceae	*Cryptocarya melanocarpa*	1			
Meliaceae	*Dysoxylum klanderi*	1			
Meliaceae	*Aglaia meridionalis*	1			
Meliaceae	*Aglaia brassii*	1			
Monimiaceae	*Tetrasynandra sp. (Mt Lewis B.P. Hyland 1053)*	1			
Monimiaceae	*Levieria acuminata*	1			
Moraceae	*Ficus congesta*	1			
Myristicaceae	*Myristica insipida*	1			
Myrsinaceae	*Rapanea subsessilis ssp. (Gordonvale S.T. Blake 9734)*	1			
Myrtaceae	*Syzygium gustavioides*	1			
Myrtaceae	*Syzygium apodophyllum*	1			
Myrtaceae	*Rhodamnia spongiosa*	1			
Myrtaceae	*Austromyrtus dallachiana*	1			
Myrtaceae	*Archirhodomyrtus beckleri*	1			
Podocarpaceae	*Podocarpus smithii*	1			
Proteaceae	*Helicia recurva*	1			
Proteaceae	*Alloxylon wickhamii*	1			
Rutaceae	*Melicope xanthoxyloides*	1			
Sapindaceae	*Mischocarpus exangulatus*	1			
Sapotaceae	*Niemeyera prunifera*	1			
Solanaceae	*Solanum torvum*	1			
Symplocaceae	*Symplocos crassiramifera*	1			
Thymeleaceae	*Lethedon setosa*	1			
Winteraceae	*Bubbia queenslandiana ssp. queenslandiana*	1			

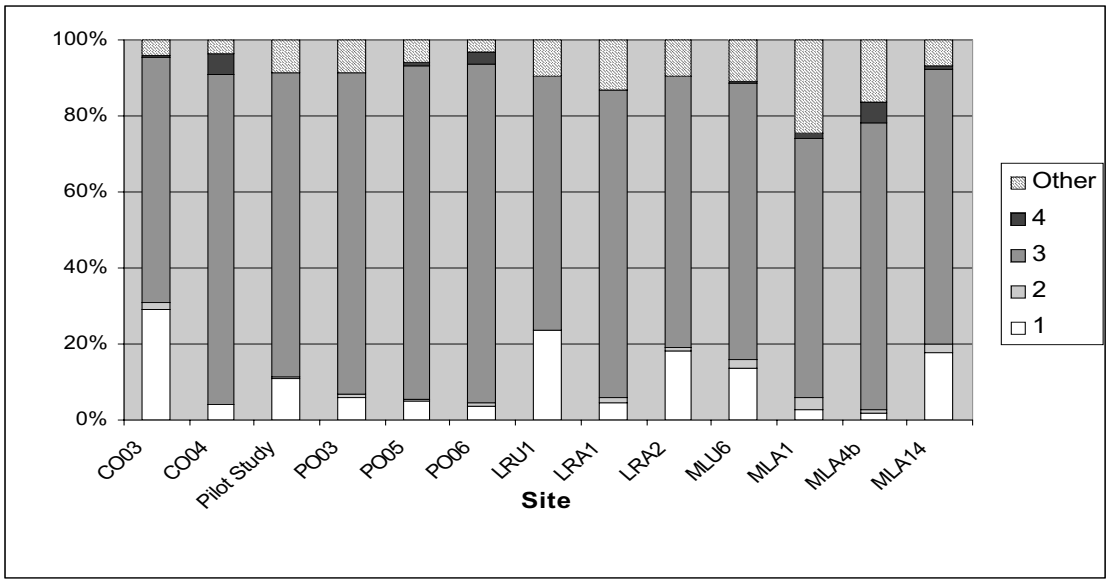


Figure 2.17. Proportion of stems at each study site falling into each successional status category. Category 1 indicates species favoured by disturbance, 2 indicates species growing in a variety of rainforest types, 3 are species of well-developed rainforest, and 4 refers to species typically growing in undisturbed rainforest.

Rare and Threatened Species

Of 316 species recorded across all sites, eighteen are listed as Rare/Threatened under State or Commonwealth legislation. The majority of listed species were recorded in the Mount Lewis area (Table 2.21), with two species in the Lamb Range area and only one recorded from the Tully Falls/Koombooloomba sites. This species, *Endiandra dichrophylla* (Lauraceae), is listed as Rare in the *Queensland Nature Conservation (Wildlife) Regulation 1994*. Although 51 stems of this species were recorded at study sites, none of these were rated as suffering from dieback.

In total, 101 stems were assessed, and of these individuals only one, *Thaleropia queenslandica* recorded at LRA1,was reported as suffering symptoms of dieback.

Threatened ecosystems

All study sites visited in the 2001-2002 surveys were within RE 7.12.16 or 7.12.1, neither of which are currently listed as “Not of Concern” in the *Queensland Vegetation Management Regulation (2000)*. However, Stanton and Stanton have mapped dieback polygons within “Of Concern” and “Endangered” Regional Ecosystems Table 2.3 Table 2.7, Table 2.15, Table 2.19)

Tree Health

With the exception of *Elaeocarpus*, no one plant genus or family appeared to be consistently affected by dieback. Eight species and two varieties of *Elaeocarpus* were encountered during surveys (Appendix II), and of these, six had an average health rating greater than one. *E. sericopetalus* had the highest average health rating of any species with more than five stems – 14 out of 29 of its stems were rated as unhealthy.

Other species consistently observed as unhealthy included *Flindersia bourjotiana* (21 unhealthy stems out of 172 stems), *Cryptocarya mackinnoniana* (18 unhealthy stems out of 85) and *Opisthiolepis heterophylla* (4 unhealthy stems out of 15).

Table 2.21. Rare and Threatened species recorded at study sites. Conservation Status, as defined under Queensland or Commonwealth legislation, is given.

Family	Species	Conser-vation Status	Recorded at sites
Arecaceae	*Linospadix microcarya*	R (Qld)	MLU6
Ebenaceae	*Diospyros sp. (Mt Lewis L.S. Smith 10107)*	R (Qld)	MLU6, MLA14
Elaeocarpaceae	*Aceratium ferrugineum*	R (Qld)	MLA5, MMA1
Gesneriaceae	*Lenbrassia australiana var. australiana*	R (Qld)	MLA14, KIA2
Lauraceae	*Endiandra dichrophylla*	R (Qld)	CO04, PO05, PO06.
Lauraceae	*Endiandra jonesii*	R (Qld)	MLA14
Lauraceae	*Endiandra phaeocarpa*	R (Qld)	MLA14, MLU6
Meliaceae	*Aglaia brassii*	R (Qld)	MLU6, MLA1, MLA14
Monimiaceae	*Tetrasynandra sp. (Mt Lewis B.P. Hyland 1053)*	R (Qld)	MLU6, MLA14
Monimiaceae	*Wilkiea wardellii*	R (Qld)	MLA1, MLA14
Myrtaceae	*Thaleropia queenslandica*	R (Qld)	LRU1, LRA1, LRA2
Proteaceae	*Helicia grayi*	R (Qld)	MLU6, MLA14
Proteaceae	*Helicia recurva*	R (Qld)	LRA2
Rubiaceae	*Cyclophyllum costatum* (formerly known as *Canthium costatum*)	V (Qld), V (Cth)	MLU6
Sapindaceae	*Sarcopteryx montana*	R (Qld)	MLA1, MLA5
Symplocaceae	*Symplocos ampulliformis*	R (Qld)	MLA5, MLA14
Symplocaceae	*Symplocos crassiramifera*	R (Qld)	MLA1
Winteraceae	*Bubbia queenslandiana ssp. queenslandiana*	R (Qld)	MLA1, MLA14

Figure 2.18 shows the number of species with at least one unhealthy stem in each of the 42 families. The worst affected families were Proteaceae, with seven out fifteen species containing at least one affected individual, and Elaeocarpaceae, with 9 out of fourteen species affected.

The list of "field susceptible" species is presented in Table 2.2. Table 2.3 shows "field resistant" species – those that are common on affected sites, and were consistently recorded as healthy. The successional status of these species showed no notable trends – the majority were category 3 species (typically growing in well-developed rainforest), with a couple of the species falling into higher or lower categories. The criteria for identifying susceptible species is deliberately conservative in that it may encompass species afflicted by pathogens or infirmities other than *Phytophthora.* For example, the list includes some pioneer species (*Polyscias murrayi* and *Alphitonia petriei*) which might be expected to have a high mortality rate. The palm *Oraniopsis appendiculata* had a significantly higher proportion of unhealthy or dead stems in unaffected sites than in affected sites (χ^2 = 11.624, df = 1, P < 0.01).

Differences in the proportion of field susceptible and field resistant species between affected and unaffected sites were significant (χ^2 = 53.55, df =2, P < 0.001). The proportion of resistant species was much higher in unaffected sites, while suceptible species were more common in affected sites (Figure 2.20). Hypothesis 6 (that species in susceptible taxonomic groups are present at lower densities in mapped dieback polygons than in unaffected forest) is not supported.

Table 2.22. Preliminary list of field susceptible tree species from all affected sites. Field susceptible species were arbitrarily chosen as those species with ≥ 5 species, and two or more of those stems showing symptoms of dieback.

Apocynaceae	*Alstonia muelleriana*	Myrtaceae	*Rhodamnia blairiana*
Araliaceae	*Polyscias murrayi*	Myrtaceae	*Syzygium kuranda*
Arecaceae	*Oraniopsis appendiculata*	Proteaceae	*Cardwellia sublimis*
Cunoniaceae	*Gillbeea adenopetala*	Proteaceae	*Darlingia darlingiana*
Elaeocarpaceae	*Elaeocarpus foveolatus*	Proteaceae	*Lomatia fraxinifolia*
Elaeocarpaceae	*Elaeocarpus sericopetalus*	Proteaceae	*Opisthiolepis heterophylla*
Elaeocarpaceae	*Sloanea australis ssp. parviflora*	Rhamnaceae	*Alphitonia whitei*
Euphorbiaceae	*Mallotus polyadenos*	Rhamnaceae	*Alphitonia petriei*
Lauraceae	*Beilschmiedia bancroftii*	Rubiaceae	*Antirhea sp. (Mt Lewis BG 5733)*
Lauraceae	*Beilschmiedia tooram*	Rutaceae	*Flindersia bourjotiana*
Lauraceae	*Cryptocarya mackinnoniana*	Sapotaceae	*Pouteria brownlessiana*
Lauraceae	*Endiandra bessaphila*	Sterculiaceae	*Franciscodendron laurifolium*
Myrsinaceae	*Rapanea achradifolia*	Xanthophyllaceae	*Xanthophyllum octandrum*

Surveys of canopy species did not include understorey early successional species, which frequently appeared as pioneers on dieback-affected sites. Resistant species in this category might include *Alpinia arctiflora* (Zingiberaceae), *Calamus moti* and *C. australis* (Arecaceae), *Dendrocnide moroides* (Urticaceae), *Gahnia sieberiana* (Cyperaceae) and *Solanum dalachii* (Solanaceae).

Table 2.23. Preliminary list of field resistant species from affected sites in the Koombooloomba/ Tully Falls area. Field resistant species were arbitrarily chosen as those with ≥ 5 stems, with all stems healthy.

Annonaceae	*Goniothalamus australis*	Lauraceae	*Endiandra montana*
Aquifoliaceae	*Sphenostemon lobosporus*	Lauraceae	*Endiandra sankeyana*
Araliaceae	*Polyscias australiana*	Lauraceae	*Endiandra wolfei*
Balanopaceae	*Balanops australiana*	Meliaceae	*Synoum muelleri*
Clusiaceae	*Garcinia sp. (Davies Creek J.G. Tracey 14745)*	Monimiaceae	*Daphnandra repandula*
Cunoniaceae	*Ceratopetalum succirubrum*	Myrtaceae	*Acmena resa*
Cunoniaceae	*Geissois biagiana*	Myrtaceae	*Austromyrtus sp. (Gillies BG 1484)*
Cunoniaceae	*Gillbeea whypallana*	Myrtaceae	*Rhodamnia blairiana*
Elaeocarpaceae	*Elaeocarpus eumundi*	Myrtaceae	*Rhodamnia sessiliflora*
Elaeocarpaceae	*Elaeocarpus largiflorens ssp. retinervis*	Myrtaceae	*Syzygium cormiflorum*
Elaeocarpaceae	*Sloanea macbrydei*	Myrtaceae	*Syzygium johnsonii*
Euphorbiaceae	*Antidesma erostre*	Myrtaceae	*Syzygium wesa*
Euphorbiaceae	*Hylandia dockrillii*	Myrtaceae	*Waterhousea unipunctata*
Euphorbiaceae	*Macaranga subdentata*	Ochnaceae	*Brackenridgea australiana*
Grossulariaceae	*Polyosma alangiacea*	Oleaceae	*Chionanthus axillaris*
Icacinaceae	*Apodytes brachystylis*	Proteaceae	*Stenocarpus sinuatus*
Icacinaceae	*Citronella smythii*	Rubiaceae	*Atractocarpus fitzalanii ssp. tenuipes*
Icacinaceae	*Irvingbaileya australis*	Rutaceae	*Brombya platynema*
Lamiaceae	*Gmelina fasciculiflora*	Rutaceae	*Flindersia pimenteliana*
Lauraceae	*Cryptocarya angulata*	Rutaceae	*Melicope elleryana*
Lauraceae	*Cryptocarya corrugata*	Sapindaceae	*Mischocarpus macrocarpus*
Lauraceae	*Cryptocarya densiflora*	Sapotaceae	*Pouteria euphlebia*
Lauraceae	*Cryptocarya leucophylla*	Sapotaceae	*Pouteria papyracea*
Lauraceae	*Cryptocarya lividula*	Sapotaceae	*Pouteria pearsoniorum*
Lauraceae	*Cryptocarya putida*	Symplocaceae	*Symplocos ampulliformis*
Lauraceae	*Endiandra dichrophylla*	Symplocaceae	*Symplocos cochinchinensis var. gittonsii*
Lauraceae	*Endiandra monothyra ssp. monothyra*	Winteraceae	*Bubbia semecarpoides*

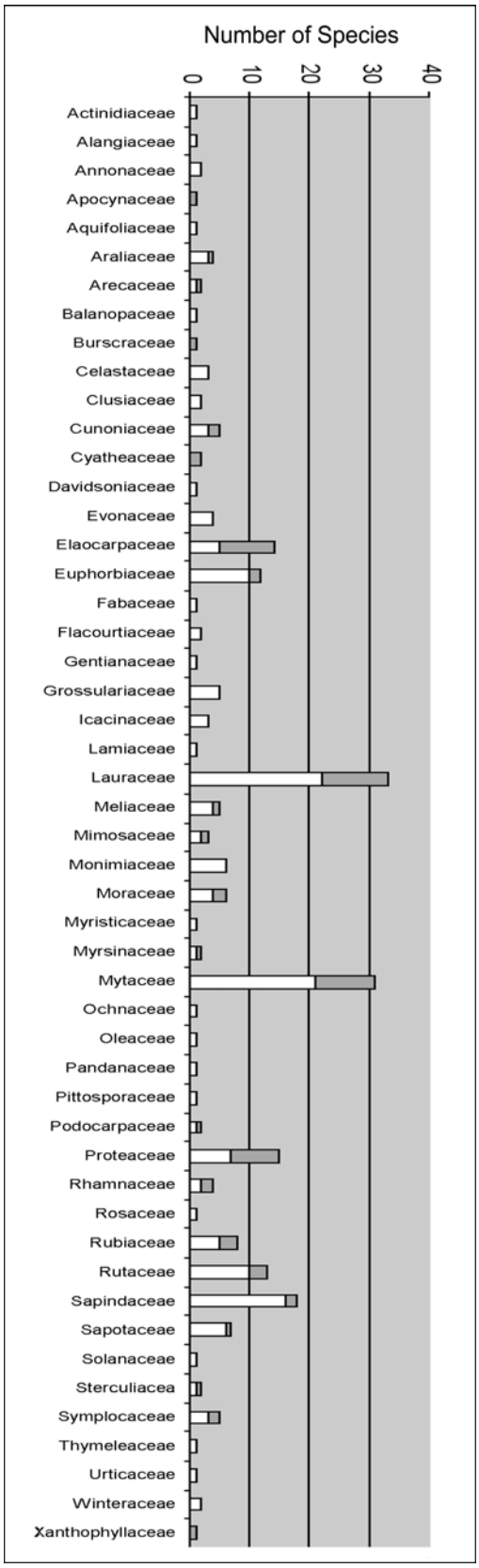


Figure 2.18. Plant families recorded during canopy surveys at field sites. Shading indicates the proportion of 'field susceptible' species in each family.

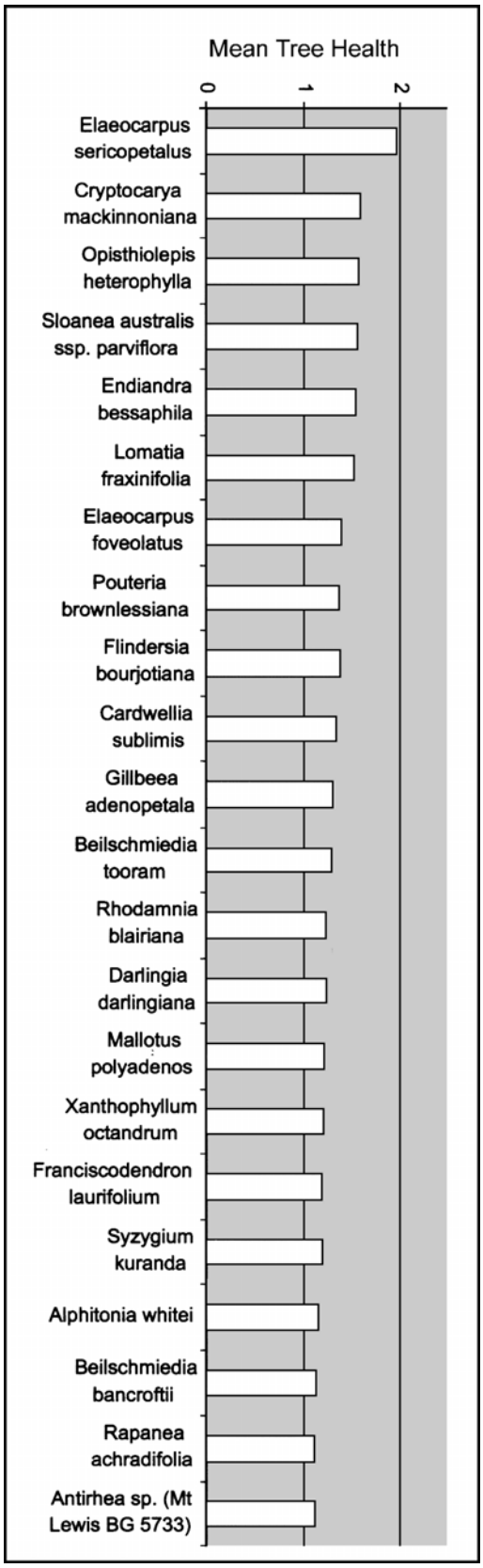


Table Figure 2.19. Mean tree health for "field susceptible" species listed in Table 2.22.

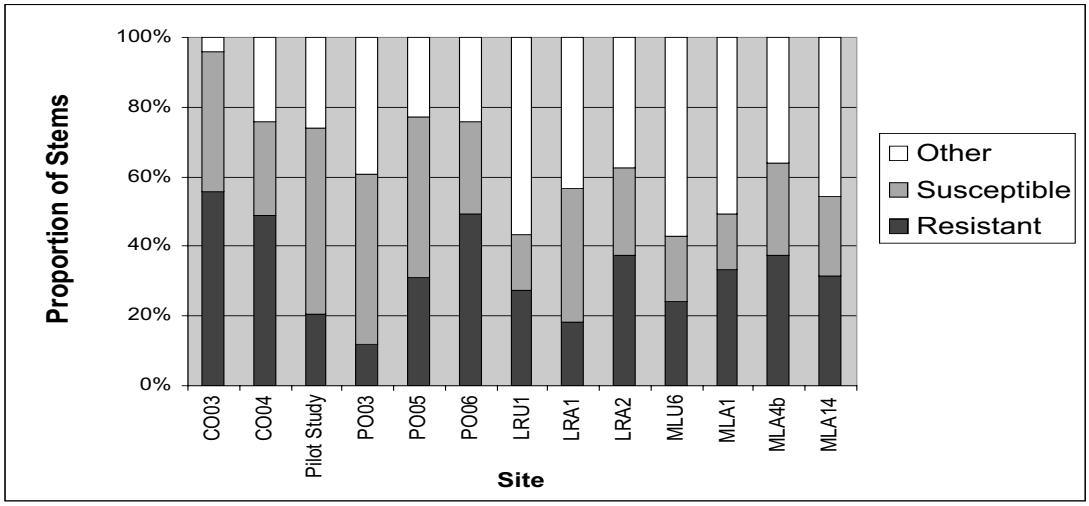


Figure 2.20. Proportion of stems belonging to field susceptible and field resistant classes (as defined in Table 2.22 and Table 2.23, respectively) present at each study site.

2.4 DISCUSSION

2.4.1 ESTABLISH PROTOCOLS TO DETERMINE DIEBACK SUSCEPTIBLE AND NON-SUSCEPTIBLE AREAS OF THE WET TROPICS WORLD HERITAGE AREA.

Protocols for determining dieback-susceptible areas in the Wet Tropics were presented in Worboys and Gadek (2002). A catchment based approach was recommended, and for each catchment, several criteria were specified for determining zones of high and moderate susceptibility to *P. cinnamomi* – related dieback. The criteria are based on analyses of Stanton and Stanton's mapping presented in Gadek *et al.* (2001), and are:

High susceptibility zones:
- notophylls dominant; and
- altitude between 750 m and 1050 m; and
- located on soils derived from acid igneous rocks.

Moderate susceptibility zones:
- mesophylls dominant; or
- rainforest has sclerophyll emergents; and
- altitude between 750 m and 1050 m; and
- located on acid igneous rocks or basalts or alluvium.

The site characteristics described above are applicable at a broad scale (1 : 100 000), and apply generally to the mapped dieback polygons of Stanton and Stanton (Figure 2.1).They were designed for landscape scale decision making, and are insufficient to describe site level physical characteristics.

2.4.2 DETERMINE WHETHER CURRENT FINDINGS APPLY ACROSS OTHER AFFECTED COMMUNITIES IN THE WET TROPICS, AND PARTICULARLY IF THE ASSOCIATION WITH DRAINAGE LINES AND ROADS HIGHLIGHTED IN THE TULLY FALLS REGION IS REPEATED IN OTHER REGIONS OF THE WET TROPICS.

Distribution of dieback across the Wet Tropics

Dieback in the upland rainforests of the Wet Tropics proved to be variable in both severity and extent. It was frequently difficult to pinpoint on the ground, even with the assistance of clearly marked aerial photography. Consequently, large, severely affected dieback patches (*i.e.* >10-15 affected trees) could not be found in either the Kirrama or Lamb Range areas. Moderately affected and small, severely affected sites were located in these areas, but large, severely affected sites were preferred as they provided opportunities for striking contrasts. The most severely affected study sites observed were located on the Carbine Tableland (Mt Mackay, Mt Lewis and the Devil's Thumb Track) although the very large dieback areas were seen in the Tully Falls/Koombooloomba area (Pilot Study site).

Impacts on floristics and forest structure

In contrast to the observations of Gadek *et al.* (2001), this study found instances of severe dieback apparently affecting most canopy species in a community. This was particularly evident at Mt Mackay, but also at MLA4b.

Many species were unique to either affected sites or to unaffected sites (Table 2.20). Although some taxa are clearly restricted to affected sites and others to unaffected sites (Appendix II), the significance of these patterns is not clear. Whilst species unique to an affected site may be present because they are better able to tolerate *Phytophthora* infection (or species unique to unaffected sites are susceptible to infection), this cannot be confirmed without a) knowledge of the susceptibility of the absent species, and b) knowledge of whether the species is recorded as absent simply because of an insufficient sampling size.

Species assessed as susceptible were significantly more abundant in affected sites than in unaffected forest (Figure 2.20). This is in contrast to the prediction that susceptible species would be quickly removed from an area, resulting in low densities of susceptible species. A community of susceptible hosts is more likely to support and promote a virulent outbreak of dieback than a community with large numbers of field resistant species. However, such communities are rarely wiped out by the infection. Weste (1994) noted that most susceptible eucalypts will continue to grow on well drained fertile soils, despite the presence of *P. cinnamomi*. Further, even if susceptible species have been wiped out by *Phytophthora*, they can subsequently regenerate on affected sites (Weste, 1997).

Position in the landscape

Worboys and Gadek (2002) noted that, although dieback was commonly observed along ridgelines in the WTWHA, analyses of the mapped dieback polygons of Stanton and Stanton found that this was not the case. There was no consistent association of mapped dieback polygons with ridgelines in the Kirrama, Lamb Range, Mount Lewis or Koombooloomba areas.

Worboys and Gadek (2002) found a significant association between mapped dieback polygons and roads in the Wet Tropics in the Mount Lewis and Kirrama areas, similar to the findings of Gillieson *et al.* (2001). This association was not significant in the Lamb Range area, however, when compared with existing 1:50 000 mapping, and with the distribution of roads observed during field surveys, the GIS layer utilised in these analyses was clearly incomplete.

Soils

In initial surveys at the Tully Falls/Koombooloomba study sites, one site characteristic did show a clear relationship with mapped dieback polygons. At all affected sites except one, an increase in clay content was detected with increasing depth through the profile (Table 2.5). Increased soil clay with depth is likely to impede drainage, which, during periods of high rainfall intensity, leads to periods of soil saturation, and/or movement of water through the surface soil layers. The stress caused by periods of hypoxia in the root zone can also predispose roots to infection by zoospores (Burgess *et al.* 1999). Where *P. cinnamomi* or other *Phytophthora* species are present, this favours *Phytophthora*-related disease by a) providing conditions suitable for a rapid build-up of inoculum (Erwin and Ribeiro 1996); and b) providing a dispersal medium for zoospores.

Although an increase in clay content with increasing depth in the soil profile was initially thought to have been associated with dieback sites, this was not borne out when investigated further at the Kirrama study sites. However, the Kirrama study sites were only moderately affected by dieback, and it may be worthwhile to apply similar methodologies in severely affected sites.

2.4.3 UNDERTAKE AERIAL SURVEYS AND MONITOR ESTABLISHED FIELD PLOTS TO TRACK THE OCCURRENCE AND EXTENT OF CANOPY DIEBACK OVER TIME.

Canopy photography

Because of the small sample sizes used (four unaffected sites and nine affected sites), canopy photography proved insufficient to distinguish between affected and unaffected forest canopies. However, canopy photography is likely to be a useful monitoring tool for interpreting progress of dieback and/or recovery, providing a consistent methodology is used. Such a methodology must:

- take into account seasonal variation in canopy openness (all photographs in this round were taken during the “wet” season)
- utilise established photopoints so that photos can be compared between monitoring events
- follow a consistent procedure for adjusting and analysing hemiphots.
-

Changes in dieback severity over time

Monitoring of the sites of Gadek *et al.* (2001) indicates some increase in the dieback severity at affected sites.

Monitoring of *P. cinnamomi*-related dieback in the Wet Tropics rainforests is problematic. In Western Australia and Victoria, its incidence and spread can be precisely measured by the health of susceptible “indicator” species in the community (*e.g.* Aberton *et al.* 1999, di Stefano 2001, Dieback Working Group 2000). In the diverse forests of the Wet Tropics, such an approach appears to be rarely applicable.

Ideally, some measurement of dieback patch size should be incorporated into the site health assessments. However, due to the diffuse nature of the symptoms at most study sites, this strategy would appear to be impractical. In some exceptional situations, (sites MLA4b and MMA1), the edges of the dieback were clearly definable, but at most, this was not possible. Where a site appeared to be recovering (sites KIA1 and MLA6), an abundance of healthy saplings further complicated dieback severity assessment.

It therefore seems most practical to retain the existing site health ratings, with some modifications:

- site drainage only needs to be assessed once
- an additional dieback severity rating is required, namely "4 – severe dieback". This would be applied where >15 trees were sick or dead, and a large proportion of understorey cover had been eliminated.
- disturbance of soil surface appears to be irrelevant
- monitoring of dieback severity in 10m x 10m plots, analogous to that conducted in this study, should continue. Tree species identification need not be continued, but tree health assessments for large saplings and trees within the study sites should be maintained.

Impacts on Threatened Species and Communities

The rare species, *Endiandra dichrophylla* was frequently observed during surveys in the Tully Falls/Koombooloomba area. This species was the equal tenth most commonly recorded species in this area, suggesting these forests are important habitat for the species. If this habitat is seriously impacted by patch death, then significant populations of *Endiandra dichrophylla* will also be impacted. Although other rare species were identified at affected study sites (Table 2.21) few appeared to be affected by dieback.

Study sites in the Tully Falls are not located in REs at threat from exposure to the pathogen. However, some of the study sites of Pryce (2000) were located on basalt soils, in RE 7.8.4, which is listed as "Of Concern".

Dieback has been mapped in tall open *Eucalyptus grandis* forest (RE 7.12.21 – listed as being "Of Concern") in the Tully Falls/Koombooloomba area. An inspection of a small area of dieback in this forest type near Mt Spurgeon concluded that *Phytophthora cinnamomi* was unlikely to be the cause. Nevertheless, other areas of dieback in this forest type should be inspected, and more soil samples taken, to confirm this observation.

The RE of greatest concern is 7.12.20 (Low microphyll rainforest on cloudy wet windswept granite highlands). This is "a habitat with a relatively high number of rare and threatened, spatially restricted and disjunct species of flora. A fragile ecosystem which has a very slow recovery period from disturbances such as trampling." (Sattler and Williams 1999). Dieback was observed (but not systematically investigated) in this and other high-altitude ecosystems during recreational bushwalking ventures in the Wet Tropics. Recommendations for further investigations of dieback in this RE are discussed below.

2.4.4 DETERMINE THE DISTRIBUTION OF P. CINNAMOMI IN THE WET TROPICS, USING THE SAMPLING STRATEGIES DEVELOPED HERE, AND DETERMINE IF P. CINNAMOMI IS LIMITED TO A PARTICULAR ENVIRONMENTAL STRATUM.

Phytophthora cinnamomi was associated with most, but not all, study sites visited in this project. Positive isolations were obtained in all areas visited – the Lamb Range, Carbine Tableland (Mount Lewis and Mount Mackay), Kirrama and Tully Falls/Koombooloomba.

Overall, the pathogen was found more often in affected sites (80% of affected study sites) than unaffected sites (67% of unaffected study sites). Similar observations of *P. cinnamomi* in apparently healthy forest were made by Brown in Gadek (1999).

Of some interest is the proportion of affected sites with negative reports of *P. cinnamomi*. There are two possible reasons for this observation:

1. inadequacy of the site selection (that is, the site was not dieback –affected, and therefore contained no *P. cinnamomi*), or

2. environmental factors causing levels of the pathogen to fall to a level that's not detectable

Site selection proved both difficult and time consuming. Accumulated errors in the process of aerial photo interpretation – GIS layer creation – mapping – onground navigation may have resulted in misplacement of study sites outside of mapped dieback polygons. Alternatively, not all mapped dieback polygons may be a result of *P. cinnamomi*-related dieback. When symptoms were moderate, they may have been confused with dieback related to other factors such as drought, lightning strike or changed drainage conditions related to road building.

It is recommended that future surveyors examine a variety of established dieback sites prior to embarking on field-based investigation. Ideal dieback patches include those at Mount Mackay (MMA1), MLA4b, PO03 as examples of severe, well defined dieback sites, and JP04, the Pilot study site as an example of more diffuse, less defined, albeit severely affected site.

3. Various environmental factors (for example, rainfall, presence/absence of a susceptible host) can influence the likelihood of isolation of *P. cinnamomi* from soils (Wilson *et al.* 2000, Weste *et al.* 2002). As 2002 was one of the driest on record in the Wet Tropics (reference), this may have influenced the frequency of isolation from study sites.

2.4.5 ESTABLISH CAUSATION (RATHER THAN INFERENCE) BETWEEN THE PRESENCE OF P. CINNAMOMI AND TREE DEATHS IN RAINFOREST COMMUNITIES, AND DETERMINE THE TRIGGERS TO VIRULENCE IN RAINFOREST ECOSYSTEMS.

Characteristic *P. cinnamomi* lesions were recorded on trees at several sites. However, attempts to isolate *P. cinnamomi* from lesions on living plants were unsuccessful.

2.5 CONCLUSIONS AND FURTHER RESEARCH

2.5.1 *P. CINNAMOMI*-RELATED DIEBACK IN THE WET TROPICS

We have demonstrated that *P. cinnamomi*-related dieback is widespread in the Wet Tropics. The pathogen is widespread, and its presence is not always related to canopy dieback, as noted by Brown in Gadek (1999). Gillieson *et al.* (2001) observed relationships between the distribution of mapped dieback polygons and roads, granite or rhyolite soils and notophyll forest type (although with a small number of incidences outside these areas). These observations have been reinforced in this study for both mapped dieback polygons and unmapped dieback patches.

The impact of *P. cinnamomi*-related dieback on some sites is dramatic and severe, in others the impacts are more diffuse and difficult to define. Although canopy openness is significantly affected by dieback, there was no consistent differences of canopy species diversity, living stem density or living stem basal area between affected and unaffected sites. However, dieback–affected stands have a higher density of field-susceptible species, which may contribute to the intensity of the disease on these sites.

In severely affected sites, it appears most species in the community are affected, however, where the outbreak is less severe, we have recorded several species which appear more susceptible to attack, and others which display a form of field resistance. Preliminary lists of field susceptible and field resistant species, based on analyses of the complete dataset from

this study, are provided in this document (Table 2.22 and Table 2.23), and supersede any lists previously distributed. It is hoped these lists will be utilised in selecting species for planting in high-susceptibility zones.

The absence of long-term monitoring data prevents us from drawing conclusions about the implications of *P. cinnamomi*-related dieback in the forests of the Wet Tropics World Heritage Area. We do not know, for instance, the characteristics of forest recovering on old patch death sites, or if the drought of 2002 affected the apparent severity of secondary symptoms associated with dieback. Although no species or communities were found to be threatened by *P. cinnamomi*-related dieback[1], the long term threats cannot be determined from the existing information. A precautionary approach to management of *P. cinnamomi* is therefore required. Management recommendations for works in high-susceptibility zones of the Wet Tropics are given in Worboys and Gadek (2002). Further monitoring and research work is required to address the following issues:

- What are the long-term impacts of *P. cinnamomi*-related dieback on forests in the Wet Tropics?
- Do soils in severely affected sites demonstrate evidence of impeded drainage in their profile?
- Establish causation (rather than inference) between the presence of *P. cinnamomi* and tree deaths in rainforest communities. This requires satisfying Koch's postulates for several rainforest tree species, as has been done in for species in southern Australia (*e.g.* Shanahan *et al.* 1996)
- Examination of regrowth in severely affected sites for evidence of selection against field-susceptible species?
-

2.5.2 DIEBACK ON HIGH-ALTITUDE RIDGELINES

A question which has arisen during the course of field work in this project is the perceived relationship between patch-death and high-altitude ridgelines (Section 2.3.3). Dieback is widespread along the western access track to the summit of Mt Bartle Frere, on the Main Range north of Mossman, near the summit of Black Mountain (Harris Peak) north of Kuranda and in the rarely visited Mt Mackay area northeast of Mt Molloy. All of these areas are in REs 7.12.19, 7.12.20 or 7.12.16. It is not known if this correlation is real, or simply an artefact of biased observations (most highland walking tracks follow ridgelines, leaving hillslopes and valleys unsampled), nor have any *Phytophthora* species been isolated from soils except for Mt Mackay. There may be some environmental factors, such as poorly drained soils or susceptible species, present along ridgelines which predispose these environments to dieback.

The observation is of significant concern. These REs are home to a high number of endemic rare and threatened species which occur in these two REs (Sattler and Williams 1999). These ridgelines are utilised by recreational walkers and land managers (and wildlife?), all of whom have the potential to spread *Phytophthora*. These areas must be considered an extremely high priority for:

- confirmation of the presence of the pathogen by soil isolations,
- establishment of monitoring programs, and
- implementation of impact mitigation measures such as user education, restriction of access to areas away from the main route and/or closure of less used tracks (in consultation with user groups).

[1] But see note on Table 2.18 regarding *Eucryphia wilkiei* on Mt Bartle Frere.

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APPENDIX I

CALENDER OF PRESENTATIONS AND PUBLICATIONS

Over the past 18 months, we have made available and publicised our research, through presentations to a variety of audiences, publications, and research reports. The following table lists some of these.

March 2003	Stuart Worboys and Paul Gadek participated in and presented at the "*Phytophthora* Technical Workshop" held by WTMA.
November 2002	Sandra Abell submitted her Honours Thesis, presenting the results of studies into the population structure and genetics of *P. cinnamomi* populations in the Mount Lewis and Koombooloomba areas.
13 August 2002	Departmental Seminar presented by Stuart Worboys, Department of Tropical Biology at James Cook University Cairns Campus. "Impacts of *Phytophthora cinnamomi* on tropical rainforest canopies."
18 July 2002	Presentation by Stuart Worboys for EV3254:03 "Tropical Agroforestry" – a 3rd year course run by the Department of TESAG at James Cook University. "Stop the Rot – *Phytophthora cinnamomi*, a significant forest pathogen."
3 July 2002	Public presentation for the Cairns and Far North Environment Centre and the Wilderness Society. "Weeds, Ferals and Dieback. The Wet Tropics Under Pressure" by Stuart Worboys
26 June 2002	Stuart Worboys led a field trip for delegates at the 3rd International Canopy Conference "Rainforest Canopy Dieback – The role of canopy science in influencing national and international environmental policies."
13 May 2002	Presentation to the Wet Tropics Management Authority by Paul Gadek
11 April 2002	Sandra Abell presented her Honours Exit Seminar "An investigation of the identification and distribution of *Phytophthora* species and the genetic population structure of *P. cinnamomi* associated with Canopy dieback within the tropical rainforests of Far North Queensland.
30 September – 5 October 2001	Stuart Worboys attended the 2nd International IUFRO Meeting on *Phytophthora* in Forests and Natural Ecosystems, held in Albany, Western Australia
September 2001	Paul Gadek convened a workshop at the 13th Biennial Plant Pathology Conference in Cairns. "Dieback in Tropical Rainforests"

APPENDIX II

SPECIES LISTS FROM ALL SITES SURVEYED.

Where canopy composition was assessed, the number of stems present at a site is presented.
Elsewhere, presence data only is given (presence indicated by "+").

Family	Species	CO03	CO04	PILOT STUDY	PO03	PO05	PO06	LRU1	LRA1	LRA2	MLU1	MLU4	MLU6	MLA1	MLA4	MLA6	MLA14	MLA4b	KIU1	KIA1	KIA2	MMA1	**Total Stems**
Actinidiaceae	*Saurauia andreana*							1		2													**3**
Alangiaceae	*Alangium villosum ssp. polyosmoides*	1			3																		**4**
Annonaceae	*Goniothalamus australis*										+		1	3	+		2	2					**8**
Annonaceae	*Meiogyne sp. (Mt Lewis L.W. Jessup 554)*					1					+				+								**1**
Apocynaceae	*Alstonia muelleriana*	2		22							+			1									**25**
Apocynaceae	*Melodinus australis*										+	+		+	+	+	+	+	+	+			
Apocynaceae	*Melodinus bacellianus*																+						
Apocynaceae	*Parsonsia latifolia*																+						
Apocynaceae	*Parsonsia straminea*																	+					
Aquifoliaceae	*Sphenostemon lobosporus*					1	2		8						+			1					**12**
Araceae	*Gymnostachys anceps*												1										
Araliaceae	*Delarbrea michieana*										+				+			+					
Araliaceae	*Mackinlaya confusa*										+	+		+	+	+							
Araliaceae	*Mackinlaya macrosciadea*			1														+					**1**
Araliaceae	*Motherwellia haplosciadea*															+							
Araliaceae	*Polyscias australiana*	1	5	20	2		2		1				1	+	+				+				**32**
Araliaceae	*Polyscias murrayi*							12	1	8			1			+	4						**26**
Araliaceae	*Polyscias purpurea*										+			1									**1**
Arecaceae	*Calamus australis*	1										+	+		+			+			+		**1**
Arecaceae	*Calamus moti*										+			+	+				+	+	+		
Arecaceae	*Laccospadix australasica*																+				+		
Arecaceae	*Linospadix apetiolata*												+				+						

Family	Species	CO03	CO04	PILOT STUDY	PO03	PO05	PO06	LRU1	LRA1	LRA2	MLU1	MLU4	MLU6	MLA1	MLA4	MLA6	MLA14	MLA4b	KIU1	KIA1	KIA2	MMA1	**Total Stems**
Arecaceae	*Linospadix microcarya*										+		+		+	+		+					
Arecaceae	*Oraniopsis appendiculata*							14	18	3	+	+		3	+	+	4	18				+	**60**
Aristolochiaceae	*Pararistolochia sparusifolia*												+					+					
Aspleniaceae	*Asplenium australasicum*												+		+		+						
Aspleniaceae	*Asplenium simplicifrons*														+								
Austrobaileyaceae	*Austrobaileya scandens*												+						+				
Balanopaceae	*Balanops australiana*										+			11									**11**
Blechnaceae	*Blechnum cartilagineum*										+	+									+		
Blechnaceae	*Pteridoblechnum neglectum*																		+	+			
Burseraceae	*Canarium muelleri*			3																			**3**
Celastraceae	*Hedraianthera porphyropetala*										+		+	1		+							**1**
Celastraceae	*Hypsophila dielsiana*								2		+	+		+		+		+					**2**
Celastraceae	*Hypsophila halleyana*										+						1						**1**
Clusiaceae	*Garcinia sp. (Davies Creek J.G. Tracey 14745)*		9	1			12				+	+	2	2	+	+	4	5	+				**35**
Clusiaceae	*Garcinia warrenii*										+			3	+								**3**
Cucurbitaceae	*Trichosanthes sp. (Mt Lewis Bgray 167)*														+								
Cunoniaceae	*Acsmithia davidsonii*																				+		
Cunoniaceae	*Caldcluvia australiensis*				2			11	3	18					+		16						**50**
Cunoniaceae	*Ceratopetalum succirubrum*							+	4	2			1										**7**
Cunoniaceae	*Geissois biagiana*									4			1				2						**7**
Cunoniaceae	*Gillbeea adenopetala*				10			1	22	3													**36**
Cunoniaceae	*Gillbeea whypallana*										+			1	+		4	2					**7**
Cunoniaceae	*Pullea stutzeri*		5	2	2			5		91							4						**109**

Family	Species	CO03	CO04	PILOT STUDY	PO03	PO05	PO06	LRU1	LRA1	LRA2	MLU1	MLU4	MLU6	MLA1	MLA4	MLA6	MLA14	MLA4b	KIU1	KIA1	KIA2	MMA1	**Total Stems**
Cyatheaceae	*Cyathea cooperi*							8		18													**26**
Cyatheaceae	*Cyathea rebeccae*		4				5	7	5	6	+			15	+		1	5	+	+	+		**48**
Davidsoniaceae	*Davidsonia pruriens*	1		1	2																		**4**
Dilleniaceae	*Tetracera nordtiana var. nordtiana*										+												
Dracaenaceae	*Cordyline cannifolia*															+	+					+	
Ebenaceae	*Diospyros cupulosa*	1																					**1**
Ebenaceae	*Diospyros pentamera*	2																					**2**
Ebenaceae	*Diospyros sp. (Millaa Millaa LWJ 515)*		1				3																**4**
Ebenaceae	*Diospyros sp. (Mt Lewis L.S. Smith 10107)*											+	1				1						**2**
Elaeocarpaceae	*Aceratium concinnum*			1																			**1**
Elaeocarpaceae	*Aceratium ferrugineum*														+			1				+	**1**
Elaeocarpaceae	*Elaeocarpus bancroftii*										+			3									**3**
Elaeocarpaceae	*Elaeocarpus elliffii*										+			1	+		3	3		+			**7**
Elaeocarpaceae	*Elaeocarpus eumundi*	1	1	5			3																**10**
Elaeocarpaceae	*Elaeocarpus foveolatus*		4	1		4		1	4	1													**15**
Elaeocarpaceae	*Elaeocarpus largiflorens ssp. largiflorens*		1	2	1			2	4	4			11										**25**
Elaeocarpaceae	*Elaeocarpus largiflorens ssp. retinervis*										+	+	+	4	+		1						**5**
Elaeocarpaceae	*Elaeocarpus ruminatus*	1						5		2													
Elaeocarpaceae	*Elaeocarpus sericopetalus*		6	1		14	6				+			1			1		+				**29**
Elaeocarpaceae	*Elaeocarpus sp. (Mt Bellenden Ker L.J. Brass 18336)*	1			1			2	2														**6**
Elaeocarpaceae	*Sloanea australis ssp. parviflora*				9			2	16	14													**41**
Elaeocarpaceae	*Sloanea langii*	3	1	1	1								5			+							**11**
Elaeocarpaceae	*Sloanea macbrydei*				2				2					1	+		10	6	+		+		**21**
Euphorbiaceae	*Aleurites rockinghamensis*	1						1															**2**

Family	Species	CO03	CO04	PILOT STUDY	PO03	PO05	PO06	LRU1	LRA1	LRA2	MLU1	MLU4	MLU6	MLA1	MLA4	MLA6	MLA14	MLA4b	KIU1	KIA1	KIA2	MMA1	**Total Stems**
Euphorbiaceae	*Antidesma erostre*			13	2		1	1			+		+										**17**
Euphorbiaceae	*Baloghia parviflora*											+			+			1					**1**
Euphorbiaceae	*Croton triacros*			8																			**8**
Euphorbiaceae	*Drypetes acuminata*		2																				**2**
Euphorbiaceae	*Drypetes iodoformis*										+			3									**3**
Euphorbiaceae	*Glochidion harveyanum*			1																			**1**
Euphorbiaceae	*Glochidion hylandii*	2						12		1					+								**15**
Euphorbiaceae	*Hylandia dockrillii*							6	3	1			+					7					**17**
Euphorbiaceae	*Macaranga inamoena*	2			1																		**3**
Euphorbiaceae	*Macaranga subdentata*										+			5	+			1					**6**
Euphorbiaceae	*Mallotus polyadenos*	16		52	4																		**72**
Fabaceae	*Castanospermum australe*	1																					**1**
Flacourtiaceae	*Casearia costulata*				1					1					+								**2**
Flacourtiaceae	*Homalium circumpinnatum*													3									**3**
Gentianaceae	*Fagraea fagraeacea*										+	+	+	1		+	+	+			+		**1**
Gesneriaceae	*Lenbrassia australiana var. australiana*										+				+		+	+			+		
Grossulariaceae	*Abrophyllum ornans*																3			+			**3**
Grossulariaceae	*Polyosma alangiacea*		3			4	6	1															**14**
Grossulariaceae	*Polyosma hirsuta*							2	1	1	+			1	+								**5**
Grossulariaceae	*Polyosma rhytophloia*	1						2	2														**5**
Grossulariaceae	*Polyosma rigidiuscula*												1		+		+						**1**
Grossulariaceae	*Polyosma sp. (Mt Lewis B.P. Hyland RFK25241)*												+					+					
Himantandraceae	*Galbulimima baccata*															+							

Family	Species	CO03	CO04	PILOT STUDY	PO03	PO05	PO06	LRU1	LRA1	LRA2	MLU1	MLU4	MLU6	MLA1	MLA4	MLA6	MLA14	MLA4b	KIU1	KIA1	KIA2	MMA1	**Total Stems**
Icacinaceae	*Apodytes brachystylis*		1			5	8	1			+		1	1	+	+	2	1			+		**20**
Icacinaceae	*Citronella smythii*				1	1		1	8		+	+		1	+			3			+		**15**
Lamiaceae	*Gmelina fasciculiflora*	2		7	1																		**10**
Lauraceae	*Beilschmiedia bancroftii*		1	2	5	3	5					+	6		+	+		+	+				**22**
Lauraceae	*Beilschmiedia collina*	1	14	8		6	7				+		5	6	+		3	2					**52**
Lauraceae	*Beilschmiedia recurva*					1		6	7				2										**16**
Lauraceae	*Beilschmiedia tooram*				10			5	18	3									+				**36**
Lauraceae	*Cinnamomum laubatii*							1	2		+					+	1						**4**
Lauraceae	*Cryptocarya angulata*		21		3	1	2	1	1	3	+			1	+		3	1					**37**
Lauraceae	*Cryptocarya cocosoides*		3																				**3**
Lauraceae	*Cryptocarya corrugata*		2					2	3	1			2		+		1	+					**11**
Lauraceae	*Cryptocarya densiflora*		7		1	9	12				+		1	7	+		+	+					**37**
Lauraceae	*Cryptocarya grandis*							1			+		5										**6**
Lauraceae	*Cryptocarya leucophylla*	1				1	2		1			+	3		+	+	1	3					**12**
Lauraceae	*Cryptocarya lividula*		4			23	11				+			5	+			4					**47**
Lauraceae	*Cryptocarya mackinnoniana*	29	1	49	6												+						**85**
Lauraceae	*Cryptocarya melanocarpa*								2														**2**
Lauraceae	*Cryptocarya murrayi*				3																+		**3**
Lauraceae	*Cryptocarya oblata*												1				1						**2**
Lauraceae	*Cryptocarya putida*		5	27		7	5												+				**44**
Lauraceae	*Cryptocarya saccharata*												1										**1**
Lauraceae	*Endiandra acuminata*												1				+						**1**
Lauraceae	*Endiandra bessaphila*				1			4	26	18		+			+		+		+	+			**49**
Lauraceae	*Endiandra dichrophylla*		15			20	16																**51**

Family	Species	CO03	CO04	PILOT STUDY	PO03	PO05	PO06	LRU1	LRA1	LRA2	MLU1	MLU4	MLU6	MLA1	MLA4	MLA6	MLA14	MLA4b	KIU1	KIA1	KIA2	MMA1	**Total Stems**
Lauraceae	*Endiandra dielsiana*			1											+			2					**3**
Lauraceae	*Endiandra hypotephra*										+			3				+					**3**
Lauraceae	*Endiandra jonesii*																+	+					
Lauraceae	*Endiandra leptodendron*									1													**1**
Lauraceae	*Endiandra monothyra ssp. monothyra*				1			1	2	3			+										**7**
Lauraceae	*Endiandra monothyra ssp. trichophylla*								1	2													**3**
Lauraceae	*Endiandra montana*		1		1		3								+		+	1					**6**
Lauraceae	*Endiandra palmerstonii*				2																		**2**
Lauraceae	*Endiandra phaeocarpa*												1				+						**1**
Lauraceae	*Endiandra sankeyana*				1				3	2													**6**
Lauraceae	*Endiandra wolfei*		3				1							4	+			1					**9**
Lauraceae	*Litsea bennettii*																	+					
Lauraceae	*Litsea connorsii*					1	2																**3**
Lauraceae	*Neolitsea dealbata*	1						4		3			4			+							**12**
Liliaceae	*Dianella atraxis*																	+					
Meliaceae	*Aglaia brassii*										+		+	2	+		+	+					**2**
Meliaceae	*Aglaia meridionalis*											+	+		+	+	1	+					**1**
Meliaceae	*Dysoxylum klanderi*						1																**1**
Meliaceae	*Dysoxylum oppositifolium*			1	2																		**3**
Meliaceae	*Dysoxylum papuanum*																	+					
Meliaceae	*Synoum muelleri*								2	4			+										**6**
Menispermaceae	*Hypserpa decumbens*										+										+		
Menispermaceae	*Stephania japonica*														+								
Mimosaceae	*Acacia celsa*	29		5						7				1			3						**45**

Family	Species	CO03	CO04	PILOT STUDY	PO03	PO05	PO06	LRU1	LRA1	LRA2	MLU1	MLU4	MLU6	MLA1	MLA4	MLA6	MLA14	MLA4b	KIU1	KIA1	KIA2	MMA1	**Total Stems**
Mimosaceae	*Archidendron grandiflorum*			2		1	1																**4**
Mimosaceae	*Archidendron vaillantii*	1		1				1			+		1	1					+				**5**
Monimiaceae	*Daphnandra repandula*							13		7													**20**
Monimiaceae	*Doryphora aromatica*				15			11	9	3							1						**39**
Monimiaceae	*Dryadodaphne sp. (Mt Lewis B.P. Hyland+ RFK 1496)*										+												
Monimiaceae	*Levieria acuminata*								2														**2**
Monimiaceae	*Palmeria scandens*										+				+		+						
Monimiaceae	*Steganthera macooraia*										+		+	10	+		+	5					**15**
Monimiaceae	*Tetrasynandra laxiflora*				1				1														**2**
Monimiaceae	*Tetrasynandra sp. (Mt Lewis B.P. Hyland 1053)*												+				1						**1**
Monimiaceae	*Wilkiea angustifolia*										+		+		+		+						
Monimiaceae	*Wilkiea wardellii*										+			2			1						**3**
Moraceae	*Ficus congesta*									1													**1**
Moraceae	*Ficus crassipes*												1										**1**
Moraceae	*Ficus leptoclada*				1			1	1	2													**5**
Moraceae	*Ficus pleurocarpa*							1	1	1													**3**
Moraceae	*Ficus watkinsiana*			1																			**1**
Moraceae	*Streblus glaber var. australianus*		1								+	+		1	+			1					**3**
Myristicaceae	*Myristica insipida*				2																		**2**
Myrsinaceae	*Ardisia brevipedata*												+				+						
Myrsinaceae	*Ardisia pachyrrhachis*										+	+		+	+	+		+					
Myrsinaceae	*Rapanea achradifolia*		6	1		8	3				+		1	2	+		+	3	+			+	**24**
Myrsinaceae	*Rapanea subsessilis ssp. (Gordonvale S.T. Blake 9734)*						1				+		+		+								**1**
Myrsinaceae	*Tapeinosperma sp. (Cedar Bay J.G. Tracey 14780)*												+										

Family	Species	CO03	CO04	PILOT STUDY	PO03	PO05	PO06	LRU1	LRA1	LRA2	MLU1	MLU4	MLU6	MLA1	MLA4	MLA6	MLA14	MLA4b	KIU1	KIA1	KIA2	MMA1	**Total Stems**
Myrtaceae	*Acmena resa*					4	1		1				+				1		+	+			**7**
Myrtaceae	*Archirhodomyrtus beckleri*									2													**2**
Myrtaceae	*Austromyrtus dallachiana*				1								+						+	+			**1**
Myrtaceae	*Austromyrtus minutiflora*			2																			**2**
Myrtaceae	*Austromyrtus shepherdii*					1									+			7					**8**
Myrtaceae	*Austromyrtus sp. (Danbulla L.S. Smith 10123)*							1	4														**5**
Myrtaceae	*Austromyrtus sp. (Gillies BG 1484)*		7			3	3																**13**
Myrtaceae	*Decaspermum humile*	1		1																			**2**
Myrtaceae	*Pilidiostigma tetramerum*	1											+										**1**
Myrtaceae	*Pilidiostigma tropicum*	4		2																			**6**
Myrtaceae	*Rhodamnia blairiana*		4	21		17	9								+		+	7	+			+	**58**
Myrtaceae	*Rhodamnia costata*									1			1				1						**3**
Myrtaceae	*Rhodamnia sessiliflora*	1		3	2																		**6**
Myrtaceae	*Rhodamnia spongiosa*				1																		**1**
Myrtaceae	*Rhodomyrtus macrocarpa*	2																					**2**
Myrtaceae	*Rhodomyrtus pervagata*			1				1		2					+			2					**6**
Myrtaceae	*Syzygium alatoramulum*							13	1														**14**
Myrtaceae	*Syzygium apodophyllum*					3						+			+			+					**3**
Myrtaceae	*Syzygium canicortex*		5																				**5**
Myrtaceae	*Syzygium cormiflorum*	1		4							+			1	+			5				+	**11**
Myrtaceae	*Syzygium endophloium*		6	3		1	2	1			+				+		1	4					**18**
Myrtaceae	*Syzygium gustavioides*																3						**3**
Myrtaceae	*Syzygium johnsonii*	1	1	6	1	3		1					1		+			1					**15**
Myrtaceae	*Syzygium kuranda*	3	9	58		3	10				+			10	+			6					**99**

Family	Species	CO03	CO04	PILOT STUDY	PO03	PO05	PO06	LRU1	LRA1	LRA2	MLU1	MLU4	MLU6	MLA1	MLA4	MLA6	MLA14	MLA4b	KIU1	KIA1	KIA2	MMA1	**Total Stems**
Myrtaceae	*Syzygium luehmannii*			1		2																	**3**
Myrtaceae	*Syzygium papyraceum*		1	13	3			1													+		**18**
Myrtaceae	*Syzygium trachyphloium*							6	7	15			1										**29**
Myrtaceae	*Syzygium wesa*		4			1	1						4		+		2	1					**13**
Myrtaceae	*Syzygium wilsonii ssp. cryptophlebium*	1									+			3	+	+		1					**5**
Myrtaceae	*Thaleropia queenslandica*							1	1	23													**25**
Myrtaceae	*Waterhousea unipunctata*			2		3																	**5**
Ochnaceae	*Brackenridgea australiana*	1	17	6		11	26	1											+				**62**
Oleaceae	*Chionanthus axillaris*		3				2		4		+	+	10	+	+	+	2	4	+	+	+		**25**
Oleaceae	*Jasminum didymum*												+			+							
Oleaceae	*Jasminum kajewskii*														+								
Pandanaceae	*Freycinetia excelsa*										+	+		+	+	+	+	+	+	+			
Pandanaceae	*Freycinetia scandens*														+								
Pandanaceae	*Pandanus monticola*										+			1	+			2					**3**
Pittosporaceae	*Auranticarpa papyracea*		1	2																			**3**
Pittosporaceae	*Pittosporum trilobum*														+	+		+					
Podocarpaceae	*Podocarpus smithii*											+			+	+		3					**3**
Podocarpaceae	*Prumnopitys ladei*												+										
Podocarpaceae	*Sundacarpus amarus*			1																			**1**
Polypodiaceae	*Drynaria rigidula*																+						
Polypodiaceae	*Platycerium bifurcatum*																	+					
Proteaceae	*Alloxylon wickhamii*									1													**1**
Proteaceae	*Athertonia diversifolia*															+							

Family	Species	CO03	CO04	PILOT STUDY	PO03	PO05	PO06	LRU1	LRA1	LRA2	MLU1	MLU4	MLU6	MLA1	MLA4	MLA6	MLA14	MLA4b	KIU1	KIA1	KIA2	MMA1	**Total Stems**
Proteaceae	*Buckinghamia celsissima*	2		13																			**15**
Proteaceae	*Cardwellia sublimis*	5	2	19	12	9	1		3	10	+		5	2	+	+	6	4		+			**78**
Proteaceae	*Carnavonia araliifolia var. araliifolia*		2			3									+								**5**
Proteaceae	*Carnavonia araliifolia var. montana*										+			7				3					**10**
Proteaceae	*Catalepidia heyana*												+										
Proteaceae	*Darlingia darlingiana*	19	7	9	5	18	10				+	+	1	2	+	+		1	+	+	+		**72**
Proteaceae	*Gevuina bleasdalei*		5			1													+				**6**
Proteaceae	*Helicia australasica*												+										
Proteaceae	*Helicia grayi*												+				+						
Proteaceae	*Helicia lewisensis*																	+					
Proteaceae	*Helicia nortoniana*	4			4																		**8**
Proteaceae	*Helicia recurva*									1					+								**1**
Proteaceae	*Lomatia fraxinifolia*			1	1	5	4			1					+		+	1					**13**
Proteaceae	*Musgravea stenostachya*								1					+			2						**3**
Proteaceae	*Opisthiolepis heterophylla*				8			1		6	+			1									**15**
Proteaceae	*Placospermum coriaceum*								1	2			+	1	+	+	+	+			+		**4**
Proteaceae	*Sphalmium racemosum*															+							
Proteaceae	*Stenocarpus reticulatus*	2																					**2**
Proteaceae	*Stenocarpus sinuatus*	2		6																			**8**
Rhamnaceae	*Alphitonia petriei*							8		35	+			1			13						**57**
Rhamnaceae	*Alphitonia whitei*	7		14	8	10	5												+				**44**
Rhamnaceae	*Emmenosperma alphitonioides*	1																					**1**
Rhamnaceae	*Schistocarpaea johnsonii*										+			2	+			1					**3**
Rosaceae	*Prunus turneriana*			1	1			2									+						**4**

Family	Species	CO03	CO04	PILOT STUDY	PO03	PO05	PO06	LRU1	LRA1	LRA2	MLU1	MLU4	MLU6	MLA1	MLA4	MLA6	MLA14	MLA4b	KIU1	KIA1	KIA2	MMA1	**Total Stems**
Rubiaceae	*Antirhea sp. (Mt Lewis BG 5733)*		17			2	7						1		+		1	8					**36**
Rubiaceae	*Antirhea tenuiflora*			2	1													+					**3**
Rubiaceae	*Atractocarpus fitzalanii ssp. tenuipes*	3	1	7	1	3	1				+		3	3	+	+							**22**
Rubiaceae	*Bobea myrtoides*		14	2		29	21														+		**66**
Rubiaceae	*Canthium costatum*												1										**1**
Rubiaceae	*Canthium sp. (Herberton Range S.F. Kajewski 1377)*	1																					**1**
Rubiaceae	*Cyclophyllum multiflorum*	1																					**1**
Rubiaceae	*Ixora sp. (North Mary L.A. BH 8618)*												3					+					**3**
Rubiaceae	*Morinda sp. (Black Leaves BGray 1677)*												+		+								
Rubiaceae	*Psychotria sp. (Daintree NP P.I.Forster+ PIF21974)*										+				+								
Rubiaceae	*Psychotria sp. (Danbulla S.T. Blake 15262)*												+				+						
Rubiaceae	*Psychotria submontana*														+								
Rubiaceae	*Psydrax lamprophylla*												+										
Rutaceae	*Acronychia acidula*	11																					**11**
Rutaceae	*Acronychia acronychioides*			1		1	2																**4**
Rutaceae	*Acronychia vestita*			2	1																		**3**
Rutaceae	*Brombya platynema*	3	20		1		6				+			17									**47**
Rutaceae	*Flindersia acuminata*	2			2																		**4**
Rutaceae	*Flindersia bourjotiana*	4	11	60	7	56	17				+	+	1	5	+			11	+				**172**
Rutaceae	*Flindersia brayleana*		1			1	1						1										**4**
Rutaceae	*Flindersia pimenteliana*	1	10	6		1	2	3			+		1	2		+	1	2	+	+			**29**
Rutaceae	*Halfordia scleroxyla*		4	2	1	13	9	1	3	2		+											**35**
Rutaceae	*Melicope broadbentiana*															+							
Rutaceae	*Melicope elleryana*							15	3	2													**20**

Family	Species	CO03	CO04	PILOT STUDY	PO03	PO05	PO06	LRU1	LRA1	LRA2	MLU1	MLU4	MLU6	MLA1	MLA4	MLA6	MLA14	MLA4b	KIU1	KIA1	KIA2	MMA1	**Total Stems**
Rutaceae	*Melicope vitiflora*				2		2	4	2	13													**23**
Rutaceae	*Melicope xanthoxyloides*				1																		**1**
Rutaceae	*Pitaviaster haplophyllus*	3	1				1				+			1									**6**
Rutaceae	*Zanthoxylum veneficum*							2															**2**
Sapindaceae	*Arytera pauciflora*							1															**1**
Sapindaceae	*Cnesmocarpon dasyantha*		1		1	1																	**3**
Sapindaceae	*Cupaniopsis flagelliformis var. flagelliformis*							1	1				+										**2**
Sapindaceae	*Guioa lasioneura*										+		1	2	+			1					**4**
Sapindaceae	*Harpullia frutescens*																+						
Sapindaceae	*Harpullia rhyticarpa*										+	+	+	+	+	+		+					
Sapindaceae	*Jagera pseudorhus var. integerrima*	1																					**1**
Sapindaceae	*Mischarytera lauteriana*		1						2	1			1										**5**
Sapindaceae	*Mischocarpus exangulatus*										+				+		1						**1**
Sapindaceae	*Mischocarpus exangulatus vel aff.*												+										
Sapindaceae	*Mischocarpus grandissimus*	1			1																		**2**
Sapindaceae	*Mischocarpus lachnocarpus*	8		3																			**11**
Sapindaceae	*Mischocarpus macrocarpus*	1		2	2					2			+				+						**7**
Sapindaceae	*Mischocarpus pyriformis ssp. pyriformis*	3																					**3**
Sapindaceae	*Rhysotoechia florulenta*												+							+			
Sapindaceae	*Rhysotoechia mortoniana*												+										
Sapindaceae	*Sarcopteryx montana*										+			2				1					**3**
Sapindaceae	*Sarcotoechia cuneata*			1																			**1**
Sapindaceae	*Sarcotoechia lanceolata*	1	3	4			15				+			2									**25**
Sapindaceae	*Sarcotoechia protracta*	1											+										**1**

Family	Species	CO03	CO04	PILOT STUDY	PO03	PO05	PO06	LRU1	LRA1	LRA2	MLU1	MLU4	MLU6	MLA1	MLA4	MLA6	MLA14	MLA4b	KIU1	KIA1	KIA2	MMA1	**Total Stems**
Sapindaceae	*Sarcotoechia villosa*										+												
Sapindaceae	*Synima cordierorum*									1			15				1						**17**
Sapindaceae	*Synima macrophylla*															+							
Sapindaceae	*Toechima erythrocarpum*	7	1		4																		**12**
Sapindaceae	*Toechima monticola*	1											+										**1**
Sapotaceae	*Niemeyera prunifera*						1																**1**
Sapotaceae	*Niemeyera sp. (Mt Lewis A.K. Irvine 1402)*												+										
Sapotaceae	*Pouteria brownlessiana*	8	1	7	6	1				1			+		+			1					**25**
Sapotaceae	*Pouteria castanosperma*	1																					**1**
Sapotaceae	*Pouteria euphlebia*		11			3	13				+		+	8									**35**
Sapotaceae	*Pouteria papyracea*					2	2	2	1						+			1					**8**
Sapotaceae	*Pouteria pearsoniorum*										+	+	1	2	+		2	9					14
Sapotaceae	*Pouteria sp. (Mt Lewis B.P. Hyland 579)*												9										**9**
Smilacaceae	*Ripogonum album*												+			+							
Smilacaceae	*Smilax glyciphylla*										+	+	+	+	+	+		+	+	+	+		
Solanaceae	*Solanum torvum*								1														**1**
Sterculiaceae	*Argyrodendron peralatum*				1								4										**5**
Sterculiaceae	*Argyrodendron sp. (Mt Haig L.S. Smith+ 14307)*												+										
Sterculiaceae	*Franciscodendron laurifolium*		3	13	7	8		19	11	2	+	+	4	10	+	+	8	6			+		**91**
Symplocaceae	*Symplocos ampulliformis*														+		1	6					**7**
Symplocaceae	*Symplocos cochinchinensis var. gittonsii*		1			5	3	1		2													**12**
Symplocaceae	*Symplocos cochinchinensis var. glaberrima*			2											+		+		+		+		**2**
Symplocaceae	*Symplocos cochinchinensis var. piliosiuscula*			2				2	1														**5**

Family	Species	CO03	CO04	PILOT STUDY	PO03	PO05	PO06	LRU1	LRA1	LRA2	MLU1	MLU4	MLU6	MLA1	MLA4	MLA6	MLA14	MLA4b	KIU1	KIA1	KIA2	MMA1	**Total Stems**
Symplocaceae	*Symplocos crassiramifera*										+			1									**1**
Thymeleaceae	*Lethedon setosa*														+			1					**1**
Unknown	*Unknown Unknown*	4	7	33	17	17	4	5	25	9			1	7			4	5					**138**
Urticaceae	*Dendrocnide moroides*							1															**1**
Vitaceae	*Cissus hypoglauca*												+			+							
Winteraceae	*Bubbia queenslandiana ssp. queenslandiana*										+			2			+						**2**
Winteraceae	*Bubbia semecarpoides*						8	1															**9**
Winteraceae	*Tasmannia membranea*																			+			
Xanthophyllaceae	*Xanthophyllum octandrum*		15	1	2	10	9				+		+		+	+	1	1					**39**
Xanthorrhoeaceae	*Romnalda grallata*																+						
Zingiberaceae	*Alpinia arctiflora*														+					+		+	
No. Species Present	**315**	**62**	**61**	**70**	**63**	**53**	**56**	**61**	**54**	**55**	**19**	**9**	**75**	**66**	**24**	**18**	**64**	**72**	**6**	**8**	**7**	**2**	

APPENDIX III

REPORT ON THE 2ND INTERNATIONAL IUFRO MEETING ON *PHYTOPHTHORA* IN FORESTS AND NATURAL ECOSYSTEMS, 30 SEPTEMBER TO 5 OCTOBER 2001.

The following summarises relevant presentations from the meeting.

Diversity of the Genus

The genus *Phytophthora* is relatively large and causes problems in crops and natural ecosystems around the world. In Australia, *P. cinnamomi* is probably the best known member of the genus, however, as this meeting made abundantly clear, it is not the only *Phytophthora* impacting on natural or managed ecosystems.

In the past few years, an increased effort in *Phytophthora* studies has led to the discover of numerous new species. The use of molecular techniques has enable construction of an evolutionary tree superior to the classification of convenience groupings described by (Stamps *et al.* 1990).

P. cinnamomi is associated with a wide range of diseases, including little leaf disease of pine, and loblolly pine decline. It was believed responsible for removal of American chestnut from the southern part of its range. Within the species, and even within a lineage, there can be a wide range in pathogenicity and morphology.

Current Research

Mark Dobrowolski reported outcomes of a survey of 780 P. *cinnamomi* isolates from across Australia and Papua New Guinea. He found that, although a high level of heterozygosity exists in this diploid organism, only a few clonal lineages were present within Australia, strong evidence that the pathogen is imported.

P. cinnamomi is widely spread, though not ubiquitous, through the south west of Western Australia. It primarily occurs where annual rainfall averages >600mm. Its distribution in New South Wales is similarly widespread, where it threatens some rare species with extinction. Susceptibility to dieback can vary widely within a family, even within a genus. Many plants do not die immediately upon infection, but persist for some time before dying.

The indirect impacts of dieback disease on small animals can be significant. Removal of cover plants (such as *Xanthorrhoea*), nectar-bearing and fruit-bearing species can lead to declines in population levels of these animals, particularly small mammals.

The presence of *Phytophthora* in natural ecosystems may invalidate predictive models for changes in plan distribution associated with climate change.

Disease Management

During the meeting, field trips took delegates to selected dieback areas. The dramatic effects of *P. cinnamomi* on these ecosystems was made clear – in some impacted areas a clear disease front advances through healthy, diverse open woodland, leaving behind a depauperate ecosystem.

In Victoria, a strategic review is underway (October 2001) of *P. cinnamomi* in Parks and reserves. It aims to map the known distribution of *P. cinnamomi*, and to develop a workable predictive mapping program, and finally to align prioriteis with those of the National Threat Abatement Plan.

Some of the methodologies identified as useful (or potentially useful) in controlling spread of *Phytophthora* inoculum included:

- nursery stock must be disease free. Use of potassium phosphonate (K_2PO_3) or other compounds as disease suppressants is not sufficient. The nursery must be completely disease free.
- in roads passing through diseased areas, removal of susceptible species reduces inoculum levels

- implementation of rigorous hygiene standards. In particular, washdown of vehicles, tools, equipment and footwear when moving between infested and uninfested areas
- dirty second-hand machinery imported from infested areas can provide a vector for disease spread
- broadscale application of phosphonate fungicides is used in Western Australia to protect critically endangered species and ecosystems. Research is underway into the long-term effectiveness of this strategy, and its impact on plant reproductive success.
- closure of roads and other access tracks
- risk assessment of proposed activities in susceptible areas
- effective mapping of the distribution of the pathogen