

PHYTOPHTHORA WORKSHOP - 15/5/80.

- 9.08. Mr. Beggs opens workshop; welcomes delegates from other states. Disease is of primary importance to the state. Effects water supplies, flora, quarantine and research program. Workshop to direct attention to the problem and give their expertise to the investigation of the problem.

Introduction of Prof. Lew Roth - A practical forester in the field of Phytophthora disease.

- 9.11. Prof. Lewis Roth

Allegory to wise old owl, who said 'think for yourself'.

Commended the application of the wide range of research carried out by some 36 researchers here. Recommends review of research over the past 10 years for possible inclusion of past knowledge.

Phytophthora cinnamomi is an international pathogen, trying to avoid reminiscing, he will try to take a wider view of the problem.

South Africa - *P.c.* present but no problem.

Europe - " " " " " - chestnuts replaced.

Japan - " " " " "

S.E. U.S.A. - " " and problem.

Little leaf disease - established on abandoned cotton fields - impermeable clay. Reached small pole or even log size before the disease observed.

Identified sites and by management contained the disease. Diversification of the species in the forest by inclusion of broadleaf spp. and resistant pines.

Hawaii - A carnage in the sp. Ohia - comparable to the Northern jarrah forest.

Auckland - *P.radiata* - little leaf disease. Added superphosphate - 18-20 days later the trees are healthy and regrowing well.

P.lateralis - *Chamaecyparis* specific - collar disease, but roots completely occupied by the fungus, spread by splash from storm water. A valuable wood, \$300 per cu. meter. Logging spread, cattle spread, roading spread, downhill spread greater than uphill.

Port Orford Cedar - If the pathogen had been an ancient introduction, the disease would not have been so devastating.

Douglas Fir - Immensely valuable forest - very susceptible as are all middle west conifer forests.

Tests of various forest soils, heated, irrigated, temperature tank tests showed disease stopped at $<15^{\circ}\text{C}$ and moisture was adequate.

Rain rapidly reduces soil temperature.

The fact that temp./soil moisture are seldom optimum in the Douglas Fir forest provides control of the disease.

Bad management in nurseries, flooding, can produce sink holes of infection; several pathogens occupying the areas.

Sanitation in the nursery is paramount.

Karri.

Ponderosa pine (Jarrah)

Port Orford Cedar (Douglas Fir forest component).

Evolution.

Plant migration.

Serial succession.

Community Development.

Over millenia, the ecosystem has established in balance, but disease can cause succession of the plant components, and the microbial population will be affected by such changes. Microbes important in nutrient cycling by degradation of the litter with leachates from leaves, etc. Provide antagonisms to pathogens.

Soil populations highly competitive. Rhizosphere population different to free living soil members; different at rhizoplane. Mycorrhizal associations act as some barrier to pathogens; all interactions are by degree and any imbalance can affect the protective ability of an organism.

Bacteria on rhizoplane highly antagonistic to pathogen infection, (major component of this population).

Degradation of the autotrophic population will deprive the soil population of food source, and this can be by removal of a minor component of a population which will affect a major component. This is true for all soil types.

Where did *P.c.* come from?

Waterhouse - *P.palmivora*.

Shepherd - *P.c.* into Australia.

Autotrophic/heterotrophic populations migrate together.

Evidence (?) - to S.E. U.S.A., Hawaii, S.Africa.

Fungus must be centred on one place.

Originally named - East Indies → Britain → Holland → Japan.

Phytophthora as a genus:

Origin in S.E. Asia - small genus with little ability to spread themselves. Spread by water, river or seas by infected root and stem material removed by storms into the sea and then migrate from shore to shore.

P.c. in mangroves in Australia.

(Ken Pegg).

Phytophthora a young genus, forebear of *P.infestans* drifted from S.E. Asia on a coconut or whatever, to spread. Infection of Hawaii by sea or man over a thousand years. This is an ongoing process.

Most of the major plant losses from Phytophthora are on spp. far removed from the first spp. from which isolated.

Overland movement by feral animals - pigs, goats, water birds by mud stuck to their bodies.

Desertification - result of removal of autotrophic/heterotrophic population. Pythium survives in such degraded areas stopping regeneration of seedlings.

Plots in forest, abandoned fields or woodland and nursery soils showed high mortality in inoculated and uninoculated plots, but disease declined only in the forest situation.

Spread by dust in high winds when saprophytic competitors have been killed by fire or whatever.

Compost - Peat moss/litter from forest floor improve exchange properties, moisture holding capacity and control disease mycorrhizal association.

Rehabilitation after a hot burn fire by encouragement of species which improve nutritional qualities of the soil.

Question:

Malacjzuk - Is jarrah forest attacked by degradation.

Roth - Requires longer observation of forest with and without fire (Went - Brazil and lateritic soils - recycling of litter so rapid that the saprophytic organisms are not included in the food chains - surface feeder possibly essential of the organic matter. Destruction of these surface roots at regular shorter intervals will degrade the ability of the trees.

Podger - Root pads for rainfall absorption - fire and removal of the litter may make the pads effective in rainfall absorption.

Roth - Agreed, but fires should be less frequent as before disturbance by modern man.

Marks - Enquired on *P.c.* on mangrove. (no comment).

Weste - Role of tolerant plants - reservoirs of inoculum? Even if they survive, and you cannot recover the pathogen.

Roth - Yes, I think so. Disease resistance.

4 types - Phytogenetic: Marri, Karri, etc.

Horizontal - Interspecific resistance not common.

Vertical - Not much chance of finding this.

Microbiological - A possible adjustment of the microbes in association with improved species tolerance.

Shearer - Microbic manipulation difficult in short term.

Roth - It will not happen quickly but in time it will. Queensland/Avocados.

Shearer - Dead zone too late for microbial improvement?

Roth - Time to reproduce a rehabilitated area, but it will take time. By introducing tolerant autotrophs to dieback areas, the soil microbial population increases.

Mulcahy - Those plants most adapted to low soil fertility are most susceptible to *P.c.*?

Havel - All components interdependent.

Roth - Agreed.

Havel - Tragedy of jarrah forest is the poor soil. Poor soils require the autotrophs to produce large root systems which are highly susceptible to root pathogens. Fertility of the soils in Vic. and W.A. make the difference between pathogen controls in Vic. and W.A.

Roth - Poorer soils require experimentation in improvement of the soil microbial population.

Mulcahy - Poor soil adaptations make the plants more susceptible to root rot.

Roth - Yes, the more delicate the balance the easier to disrupt.

Malacjzuk - Mycorrhizae - Banksia generally non-associations mycorrhizae.

10.20. Session ends.

11.05. G. Weste - Victorian situation.

Pathogen must be present.

Abiotic and Biotic factors.

Temperature - Soil Temp. $<10^{\circ}\text{C}$. Little activity.

Sporangia - 20°C . Optimum activity.

Depression in baiting in cold weather.

Winter population drops.

When soil moisture low, pathogen population drops.

Light - no effect.

Soil depth - profound effect - Root drainage especially - not slope as much as depth. Shallow soils - larger populations, especially on impervious layer with poor drainage.

Zoospores depend on free water for dispersion.

Chlamydospores (?) depend on particle size for survival.

$>4\mu\text{m}$ diameter in loose fabric soil pores.

Aeration and drainage.

Soil moisture content - Water potential important in comparison of soils of various kinds.

Curves for wetting up and drying up.

Brisbane Ranges - 2½% soil moisture difference between optimum and saturation.

Total soil water potential.

Osmotic potential - Matric potential.

P.c. is sensitive to water stress, not salts.

Adding amendments changes microflora.

Mycelium -

5 - 1500

50 red - 2000 Kpa - 2500.

>-500

- 1000 - 2000

Sporangia -

-80

- -160

Chlamydospores -

1050 - 2950

- 3000 Weste.

Oospores -

2950

- 3650 (*Trichoderma viridis*)

Hyphae -

> -4000

Seasonal effect:

No zoospores in summer; few in winter, many in spring. Free water for zoospores to infect. Water potential 0.1 Kpa effects zoospores.

Free water for a period to allow sporangial development and release of zoospores.

Low oxygen increased exudates - increase infection by zoospores.

Adaptability of *P.c.*

Free water - Zoospores.

Drying up of soil - Chlamydospores autumn main population - built for survival at different water potentials, -5 bars soil potential - survival to -3000 Kpa.

In Victoria, roading spreads soil infected with chlamydospores downhill and in culverts. Collected from a *P.c.* dead zone.

Drought

Predisposes host. Hosts die off due to drought and behave as though they have a nutrient deficiency. Despite reduction in transpiration, they cannot cope.

Soil Texture

Sandy soil - light rain - zoospores produce.

Clays & Loam - less zoospore production.

Gravels & sands - 86% disease severity.

Loam - 50% " "

Mulches - Increase microbes - inhibits *P.c.*

Exudates - pH - very low 3.8 - 3.6 - not practical.

Phosphorous

11.30. Discussion.

Grant - Is temperature more adverse to host as well as *P.c.*

Bumbieris - Yes, found in S.A.

Palzer

Podger - 15-18°C probably optimum.

Shea - Soil temp./moisture regimes and soil moisture have not been sorted out.

Roth - A lot of fungal activity below 15°C. but infection optimum at 15-18°C. Host may be producing more roots for infection.

Grant (?) - Tropical soil temp. higher than optimum temperature. Enzyme - temperature sensitive.

Marks - Long term survival of pathogen, in a highly infected site - antagonism greater in winter.

Bumbieris - Sterile conditions - sporangia 25°C.

Non sterile conditions - sporangia 15°C.

Palzer - No threshold for infection in time.

Gerretson-Cornell - Any information on oospores germination, etc.

Weste - No, they do not appear to be important, except as survival.

Shea - Temperature and host root growth especially in jarrah coincide with high fungus activity.

Podger - Support Chris that infection in time was not temperature tied.

Shea/Palzer - (Contretemps) - Summer rain increases root growth in jarrah.

Threshold of resting spores on temperature?

Palzer - Yes, temperature/survival in Tasmania evident.

Shea - A few degrees difference could make the disease more evident.

Roth - No disease below 15°C was in his area but may not hold elsewhere.
P. palmivora was tropical but is now progressing to cooler areas.
 Phytophthora are adapting as time passes, to cooler locations.

Marks - Stem lesion deaths - pathogenicity of Phytophthora strains from isolates from cool areas more aggressive at that temp.

Roth - Frost no barrier to infection in some strains.

Soil Depth & Drainage

Shea - Massive disease in 10 metres in W.A.

Weste - Yes, but depth delays affect.

Shea - Yes, but *P.c.* will be active in free drain site.

Weste - Agreed.

Dell - How deep in lateritic soils can *P.c.* be isolated?

Weste - Krasnozem and deep soils in root areas.

Podger - Roots, water, Oxygen, some *P.c.* damage - depth not important.

Shea - Top 4cm in jarrah. I do not think *P.c.* moves through most soils.

Dell - Why do you think it does not operate vertically.

Shea - If host supports *P.c.* will spread, but more resistant spp. do not appear to become infected at lower and larger roots.

Weste - Roots and aeration - *P.c.* follows.

Roth - Do you not get Banksia roots at surface.

Shea - Infection in Banksia and other situations overland.

Water Potential.

Marks - Mortality of zoospore and release 1000 bar.

Weste - Surface motility important in zoospore release at low water potential.

Shea -

Marks - Root growth in tall containers with soil of same particle size with water table known - roots survival good at aerated area - infection in flooded area.

Palzer - Sterne and Zentmyer - 2.5 bars and higher for infection not practical.

Physiology of the sporangia - zoospore release through 8µ. Zoospores move through soil as amoeboid motion.

Shea - Is not release activity different to soil movement.

Palzer - Not in my experience.

Old - Amoeboid movement through soil - I agree.

Bumbieris - Zoospores remain more active in free water conditions.

Podger -

Palzer -

Shea - Soil moisture not static - fluctuating more important and not static, as Geoff Marks technique.

Marks - My demonstration only a model for infection process. Allan Smith has shown that microbes on soil crumbs produce ethylene - antagonistic to pathogens. The wetting process in soils and krasnozem could accent the infection process in these soils.

- Roth - What about where litter or no soil is present?
- Marks - Tried isolating under clay over sand but was unsuccessful.
- Weste - Same under clay pan in Brisbane Ranges.
- Gerretson-Cornell - Agreed. Example, Sunny Corner - 45 pine show little leaf - only found *Phytophthora* in the surrounds.
- Shea - Why chlamydospores increase population at low soil levels at 5 Kpa.
- Weste - Where no organic content, no chlamydospores.
Even minimum organic content, positive chlamydospores.
Multiply in saprophytic fashion on the said organic material via sporangial production.
- Shea - This is important for management of forest to stop spread of the fungus by chlamydospores where organic material.
- Roth - Propagule absorption from the organic matter requires more time.
- Weste - Experience proves it happens.
- Marks - Kassaby - chlamydospores germinate, produce zoospores and mini chlamydospores in the hyphal extensions.
- Palzer - Spores of hyperparasites?
- Old - I shall present supporting evidence of mini chlamydospores.
- Gerretson-Cornell - 2 out of 320 samples from gravel positive *P.c.*
Sufficient inoculum.
- Weste - -
- Podger - Gravel should be defined. Varies from the A horizon of the soil to crushed rock, which cannot support organisms.
- Shea - Spread down rivers in South Africa.
- Weste - Yes. Outlined von Broembsen story.
- Weste - Is there a reservoir of inoculum in tolerant spp.
- Gerretson-Cornell - They found *P.c.* on resistant spp.
- Podger - Not a very productive research project.

Soil Texture.

- Havel - Fertility and soil testing.
- Marks - Why do Krasnozems and sand of similar pore size behave no different in infection response. Is it not texture/structure rather than pore size. Nurseries damage soil texture increasing disease potential.
- Weste - Clay particles important for exchange and microbial population.
- Shea - Suspect a lot of pot trials irrelevant due to unsimilarity to the field conditions. Background measurements essential.
- Weste - Pots to field.
- Shea - No, I think Dunniway techniques should be used.
- Marks - A lot more attention should be paid to structure of the pot trial soil and even the size of the pot.
- Weste - Thanks. 12.30. close.

Lunch.

- 13.30. Podger - Thanked Dieback Fund and Dr. Shea for arranging his visit. 20 years since he started *P.c.* investigation. 15 years since *P.c.* first reported at Toowoomba Conference. Long time since he worked on *P.c.* but he has maintained an interest.

Pure research issues should not be put forward as palliative management techniques until their practicality has been proved.

The Pathogen.

Pathogenicity of isolates - is there any real variation?

Marks - I disagree that the pathogen is uniform.

Heather - Difficult to test and analyse.

Old - Variability in the host make studies of pathogenic variation difficult. Perhaps *P.c.* is so variable that it can cause disease in a variety of conditions.

Bumbieris - Culture can lose virulence.

Gerretson-Cornell - I found different reactions.

Marks - The methodology for testing pathogenetic differences is not good enough due to host variabilities.

Roth - Methods available not being used. Is the A2 more pathogenic than the A¹.

? - I am not convinced there is a difference between A¹ and A2.

Roth - Why does one have a wider host range than the other.

Podger - Have you a sound basis for saying that the A2 is more pathogenic than the A1, except for experimental tests using mostly A2 isolates.

Roth - Yes, this has not been studied sufficiently.

Podger - Immediate work on A1/A2 affirmed.

Roth - Differences of pathogenicity.

Hosts and Situation Variability.

Podger - With mixed stands, approach must be in line with resistance. Jarrah suggests biological control is possible.

Manipulation of the stand by eliminating more susceptible species a practical management technique.

Marks - Host reaction; how can a jarrah collapse with a good crown in non-aggressive climates.

Podger - How about toxic leachates, barrier species, grasses.

Shea - *Acacia pulchella* has shown in pot trials that some control can be provided, but this needs to be confirmed in the field.

Malaczuk - *Poria* behaves quite differently to *P.c.* and parallels on this work were not applicable.

Roth - Production of nitrate rather than toxic substances.

Marks - Could moisture control with lower temperatures not be more effective.

Podger - *E.stringens* suppresses all other spp.

Kagi - Epoxide or Peroxide produced by this spp.

Roth - Is this only in anaerobic conditions.

- Dell - Timing of allelopathic compounds released in the environment important.
A water soluble allelopathic compound more likely to be effective.
- Shepherd - *Acacia pulchella* is hoped to produce such compounds in the jarrah forest.
- Mulcahy - Where would you look first?
- Podger - This would need thought.

14.05. Suppressive soils, rhizospheres.

- Podger - Rhizosphere a different world to the soil. Definition of suppressive and conducive soils.
- Conducive soil not a good term because it suggests a lack of antagonists.
Krasnozems take more wetting up to field capacity, compared to a laterite.
More plant growth on Krasnozems, less rain reaches the soil.
- Suppressive soils are different by their physiology as well as their other features.
- Weste - *E. obliqua* on different sites, died, but *P.c.* disappeared in the well drained warmer sites.
- Podger - Asked Chris Palzer - what about suppressive dolomite soils in Tasmania.
- Palzer - Axenic trials of zoospore inoculum showed that with sterile spores compared to Krasnozems or laterite, there was little difference on infection.
- Malaczuk - Sterilised Krasnozems behaved conducively whereas sterilised laterite was antagonistic.
- Old - -
- Palzer - Host, lupins and jarrah aseptic rhizosphere with axenic cysts; there was very little difference at the infection court.
- Malaczuk - Rhizosphere is important, also rhizoplane.
- Old - Experimental evidence - roots colonised and placed in Krasnozems and lateritic gravel; survival better in latter. Probably difference at point of survival rather than infection.
- Podger - Summarised and passed to.....
- Shea - Some of the organisms tried that have been involved in this suppressiveness - clue to relevance to control.
- We must look at *P.c.* as a pathogen and as a food source for other soil microbes.
Bacterial associations - are there any breakthroughs.
- Marks - Trying to alter suppressive soils by 1) break-up of structure, 2) removal of microbial population will destroy suppressive effects.
A soil survey in Victoria by geologists.
- Shea - Is there a suite of bacteria that are necessary for sporulation.
- Gerretson-Cornell - Not only bacteria, antibiotics produced by other than bacteria.
Starvation of the microflora generally more important. Study debilitated site by gradually adding to the population.
- Roth - Isn't there a specific group of organisms that are lytic.
- Malaczuk - No, we could not identify significant differences between single organisms - generally there was a suite of bacteria involved.
No work has been done on these suppressive or conducive organisms.
- Gerretson-Cornell - The microbial population changes with environmental changes.
- Titze - Cross protection by fungi - *Pythium irregulare* provided a cross protection of about 30% even in Banksia.
- Podger - Could this be a hyperparasite of *P.c.*
- Titze - Not investigated.

Animals - Microfauna.

Podger - Microfauna - nematodes.

Old - Amoeba and their mechanism of destruction. Predatory Amoeba have been found widely in Australia but not in lateritic soils of W.A. but have in the Krasnozem, probably due to increased microbial population supporting the Amoeba.

Podger - Can these Amoeba control *P.c.*

Old - They could, to a small extent, but I do not know enough of the biology of predatory Amoeba here.

Roth - Do we accept hyperparasites as a biological control of *P.c.*

Podger - Yes, I accept it.

Shea - Soil microbial population is important, especially where *P.c.* is continually active, but must be at maximum effect in W.A. at a very short period in time.

Malaczuk - When the burst occurs in both *P.c.* and the antagonist needs to be determined.

Shea - Due to large root inoculum in jarrah forest, soil microbes could not cope.

Weste - Antagonists not active when *P.c.* is at maximum operation.

Malaczuk - Have not seen holes in chlamydospores.

Old - No difference in chlamydospores survival in lateritic gravel compared to coastal sands.

Podger - Biological control - control of fungus and control of disease may be quite different things.

The host is being ignored in preference to the pathogen - maybe improvement of host conditions could provide control.

Why incorporate litter throughout a soil when not natural.

Malaczuk - Pot experiment with seedlings simulate the root zones in the field.

Podger - I think the litter should be applied to surface.

Malaczuk - By adding litter to soil, increased microbial activity.

Roth - A seedling's condition suggests that the litter should be at the bottom of the pot. Seedlings push roots down - mature plants up.

Mycorrhizae.

Bumbieris - Pines were protected in U.S.A. with mycorrhizae. Described trial with sterilised and natural soil inoculum, and not inoculated with *P.c.* Autoclaving increased infection and death by *P.c.* in the order of 1 - 10. The symbiont was not determined.

Boughton - I have not been able to determine any effect of mycorrhizal roots in reducing infection by *P.c.*

Marks - Mycorrhizae probably not involved in protection because the mycorrhizae in Eucalypts are found near soil level, where *P.c.* infects the roots.

Podger - Mycorrhizae not effective against root necrosis, but could be effective on feeder root necrosis.

Shea - Abundant mycorrhizae to be found on jarrah feeder root pads but infection occurs behind the feeder roots.

Dell - 12 months onwards for answer.

Roth - A good mycorrhizal association is essential for protection against infection.

Malaczuk - Amphion block.

Root activity out of the mineral soil into the litter area where the

mycorrhizae are also found. Most are basidiomycetes; fine roots are quickly occupied by mycorrhizal fungi.

With no organic matter the same mycorrhizal association is not be found, except for *Cennocucum* spp. with a small effect.

Ecto mycorrhizae depend heavily on organic material and continuous fire will reduce them.

Podger - Microfauna work being done at W.A.I.T.

Marks - Forest Commission - Victoria - 140,000 hectares reduced the site factor to 8 points.

Shea - Nick Malaczuk seems to suggest no burning.

Malaczuk - No. Freshly produced ash and litter most active mycorrhizae.

Podger - You can't afford to have 10 years of litter in W.A. due to fire catastrophies, and cannot be a practical option. Therefore Acacia/litter regimes requiring no burn would be unacceptable.

Malaczuk - But mycorrhizae most active in ash and litter.

Techniques - Stimulation of sporangia production.

Malaczuk - Hwang - cellophane sheet on agar, only suitable to laboratory trials.
Soil extracts 1:2 with low populations.
1:10 with high populations.

Stan ? - Banksia leaves 4mm in diameter inoculated, placed in the field and recovered, provides even mycelial cover around the edge of the leaf.

Darling - I commend the use of Nuclepore membranes.

Gerretson-Cornell - Cotyledon of *E. sieberi* - surface sterilise in 70% Ethanol.

Detection of *P.c.*

Shea - Hymexazol - great aid.

Shearer - Control of Pythium not 100% - Masago - 15cm dishes. Recycling plates - washed - cold sterilised with propylene oxide - wet tissues in bag - 5-10ml propylene oxide in vial, opened in bag when humidity high.

Weste - No random sampling due to host specificity of the pathogen.

Hopkins - Not only detection required but quantitative could also.

Podger - Manager only requires to know positive and negative.

Shea - Forest Research quantitative test required. Bias of isolation technique favours *P.c.* Elaine Davison to look into other species.
Chlamydospores probably germinate and produce sporangia.

Weste - 12 hrs. maximum period before observation.

Zoospores - Methods.

Byrt - Acid washed glassware. 2M acid. Pipette slowly with wide bore.
Tissue culture trays - 2ml per tray, fix with gluteraldehyde, examine with inverted microscope.

Palzer - Concurs that zoospores are long lived as cysts with micro papillae; will survive at least 6 weeks at 5°C. Axenic cultures will all germinate.

Fluorescent Stain and Brighteners.

Malaczuk - Fluorescent antibody techniques - Dunniway - antisera against *P.c.*
Cytoplasm/cell wall separated - cytoplasm into rabbit - bleed rabbit and coagulate to specific antisera.

Weste - Sterilise calcafluor, add to media (200ppm) before set. Grow the fungus.

Root Manipulation Techniques.

Old - Surface sterilise seed to produce roots - infect and place in membrane sandwich 0.2μ , 5.0μ and 200μ , sealed with vaseline circles. Incubate in soil.

Half roots are plated and half placed on slides.

Palzer - F. Podger isolated from large roots from natural infection. Excavate roots, place in vermiculite and grow various root types and apply zoospore suspension with known dose.

Marks - Remove very small area of surface bark, place mycelium cover with moist cotton wool. Infection can be measured by lesion, aspect differences by several locations. Deeper inoculum produce kino reaction.

Sites - Physiological reaction of spp. - seasonal variation.
Host pathogen relation.

Podger - We should inoculate in sites that are relevant to root rot resistance. For pathogenicity trials - use of massive spore inoculation doubtful, but host tissue should be used as inoculum which is more realistic and *P.c.* will survive for long periods.

Boughton - Pine branches from young trees - remove bark - inoculate with mycelium - 2-3 weeks completely colonised - in moist soil, lasts up to 6 months in pots.

Marks - Eucalypts not good due to kino production.

Shea - Banksia in good supply.

16.40. Posters Discussion.

16th May, 1980.

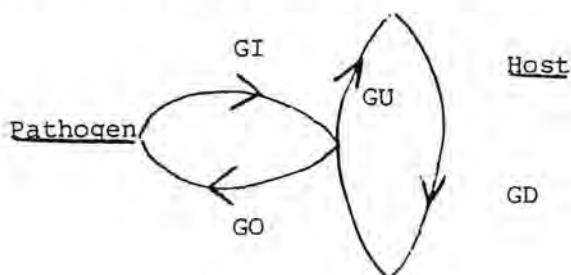
Host-Pathogen Interaction - Chris Palzer.

- 09.05. Two previous sessions have been a primer to the host-pathogen interaction at the infection court.

Reasons for research into host-pathogen interactions:

Gene frequency - screening of species for pathogenicity.
National Parks & Forests - under severe conditions for the host.
Mechanisms of infection.
Precursor to control methods.
Sources of genetic resistance for replacement species.

Model for host-pathogen interaction research



Going in - After infection of the host in the rhizoplane by one or other of the tactic responses. A trapping effect by the roots with encystment caused by root exudates.

Hinch - The role of receptors on the spores with particular reference to lectins.

Old - How do the polysaccharides relate to the cell walls.

Hinch - Similar but with added sugars.

Old - Monocotyledons produce copious mucigel; is this true of dicotyledons, with or without mycorrhizae.

Hinch - True, have not worked on mycorrhizae; worked mostly on monocotyledons, especially little work on Australian natives. Bacteria important part of the studies.

Byrt - When the zoospores approach a plant, various things occur - I would distinguish between trapping and sticking. Something else will determine germination. Trapping not caused by monosaccharides. Zoospores of *P.c.* less sensitive to monosaccharide compounds than other *Phytophthora* spp. Proteins more effective in *P.c.*

Palzer - Do you see any scope for manipulation of the mucigel layer.

Hinch - I would be hesitant to advocate this due to all the other factors involved; mycorrhizae, bacteria, etc.

Palzer - Clay particles in soil also affect the spores.

Marks - Pauline Byrt and Jill Hinch's investigations probably sequential.

Palzer - Yes.

Other spore forms also cause infections.

Chlamydospores can germinate in a day and cause infection.

Podger - Detection of chlamydospores, techniques found similar, and sensitive. 1 chlamydospore in 50g soil.

Palzer - Chlamydospores static, with root exudate stimulating germination, could be either by germ tubes or by zoospores following sporangial production.

Davison - Hyphae should be considered more, maybe the infection period could be extended by this method.

Weste - By adding mycelium to soil, disease was obtained, and could be measured. Uphill infection by mycelium in Victoria much more likely due to lack of moisture than negative geotaxis of zoospores.

Bumbieris - Mycelium in soil not found.

Weste - No, it is in the roots and spreads from root to root.

Shea - Lew Roth, Zentmyer, suggested uphill infection was by mycelium.

Roth - Rich fertile soil hyphal growth modest; perhaps more extensive in poor soils. Lot more surface activity in the disease. Possibility of splash dispersal should be considered.

Weste - We did demonstrate mycelial growth through soil and gravel. Brisbane Ranges more likely root to root infection.

Roth - Internal root growth or multiple root infection.

Weste - I don't think we know.

Bumbieris - Growth depends on nutrient availability and antagonism by microbes.

Old - On excised roots, in laterite soil, considerable growth around the roots, behaved as saprophyte in the trial conditions.

Palzer - By adding massive food sources misleading results may be obtained on *P.c.*

Wallace - Other organisms that live in the soil? A seething activity of microbes in the soil must move the fungus uphill.

Shea - If these organisms were doing this, the disease would be greater in W.A.

Palzer - Yes, but the method must be studied.

Palzer - Not every spore will infect. Probability of infection a very useful single index. Van der Plank says, often exaggerated doses produce quite artificial host interactions. I agree with this and much smaller dosages of zoospores should be used.

Byrt - 1 or 2 zoospores on a root tip would cause an infection. By dipping seedlings in zoospore suspension, infection only occurs on some of the roots - diversity of root states.

Palzer - Initially small doses of inoculum should be applied at the infection court.

Byrt - Determine infection first.

Palzer - This supports the probability of infection index.

Heather - Difficult to determine the field infection dose.

Palzer - Incubation periods and growth patterns will be important.

Morphology of Penetration.

Palzer - Appressoria not reported for axenic conditions, but positive appressoria have been seen. Hypersensitive reaction in *Acacia pulchella*.

Marks - At the point of infection by a zoospore, the germ tube enters the host and the spore forms a mycelial plug at the neck of the hyphal germ tube. High osmotic pressure?

Hinch - Calose formation, to inhibit further infection.

Gerretson-Cornell - How much protoplasm left in the sealed-off cyst.

Marks - Less than 100th of the original contents.

Palzer)
Old) - Yes, a protective mechanism.

Byrt - In low nutrient systems, all the protoplasm will move along the germ tube.

Marks - The speed of formation was surprising. Axenic conditions, observed under E.M.

Palzer - The morphology remarkably similar in a range of hosts.

Malaczuk - Cells of host collapse followed by hyphal extension. Spore dosages important in host response.
Once infection occurs, the plug develops.

Palzer - Biosphere - Microbes of various families, genus, etc.

Micro organisms in the rhizosphere have a huge effect on infection.

Boughton - Important at infection but not at later stages.

Bumbieris - Phytophthora a root infection, not collar.

Malaczuk - Micro organisms are very important in competition for exudates from the host by forming a barrier and reducing the ability of Phytophthora to infect.

Palzer - A fact, that mycorrhizae forms a physical barrier to infection, and mycorrhizal exudates also anti *P.c.* Bacteria relatively few numbers on the root and could be, but not proven, as affecting infection by *P.c.*

Malaczuk - This is a simplistic view. In soil there is a mixture of organisms of which bacteria are a component.

Palzer - This method of determination of inoculum potential for host described. 7.5 reduction of infection in non-sterile compared to sterile situation.

Bumbieris - Has anybody looked at forest trees to see if there are bacteria on the roots.

Malaczuk - Affirmed.

Shea - Boughton found plenty of mycorrhizae on *P.radiata* roots - trees still dying, and on jarrah trees also.

Malaczuk - Your jarrah observations were after fire, with fresh organic material.

Palzer - Mycorrhizae have been documented as being more effective than any other organisms.

Old - All parameters will affect the infection and survival of the fungus. Soil microbiology important as possible method of manipulation of the disease infection area.

Palzer - Yes, but the components must be investigated by factors.

Wallace - I think they should be investigated simultaneously.

Host Physiology.

Shea - Physiological differences caused by environment.

Weste - Zentmyer found site differences significant in infection.

Peter ? - Isopogon infected with 100-200 chlamydospores soon die, probably due (Vic. Del.) to water reduction. Stomatal measurements indicated water stress in Eucalypts.

Palzer - Foster and ? from Ireland showed with ? *ellwoodii*, with low infection doses they obtained different responses when the host was not producing new roots; i.e. related to day length.

Boughton - What condition were they growing in. As roots extend, they grow out of the mycorrhizae.

Wallace ?- Searle and Wilson - apple trees more susceptible to infection in spring in England.

Weste - Do you have a summer growth burst as in South Australia.

Shea - In jarrah we do have a peculiar growth of shoots in December.

Weste - South Australian summer growth after spring flowering.

Havel - Speculation.

Weste - Not speculation; years of evidence. Shortage of nutrients.

Palzer - Shigo showed that just before leaf flush, the host was particularly susceptible to disease.

10.35. Close - tea.

11.05. Colonisation - Lesion limitation - Disease Expression.

Palzer - Colonisation after infection has occurred.

Various colonising effects on different hosts.

Jarrah gradual decline, whereas total colonising of Banksia.

Cahill - Breakdown of pathogen root cells by the fungus with massive leakage of exudates - electrolyte calcium, KCl, massive change in cell physiology, which acts as food base for the fungus.

No evidence in Phytophthora that a toxin is produced, but there may be.

Marks - 1-6 -beta-glucans 1.5mg small *E.sieberi* seedlings dipped in toxin and nutrient solution, showed cell damage in 1 hour.

Palzer - Halsall also demonstrated wilting and plasmolysis with fungal cytoplasm, also Joanna Tippet.

Hart - Represented her results on cyanogenic glycosides and P.c. Much more research into which cyanogenic glycosides are present in which plants and quantity required for breakdown to hydrogen cyanide.

Malaczuk - Is this a volatile or a solid?

Hart - Could be both. Resistant and non-resistant plants, flush of spring growth.

Podger - Toxin translocation - important question. NO way consistent with a systemic toxin.

Palzer - It is only suggested that there is only one cell ahead of infection degradation of cyanogenic glycosides to hydrogen cyanide not a systemic affect.

Marks - Kino formation could be response to such a toxin.

Shepherd - At W.A.I.T. this work will be continued. Macadamia does contain cyanogenic glycosides and is highly susceptible to P.c.

Palzer - Macadamia, walnuts, etc. infected in different areas - stem, twig, etc. Type of inoculation again important, with small doses infection stops before suberisation of the root; heavy doses will alter host response due to gross infection.

Host physiology after infection largely unknown.

Marks - Increase in resistance to water conduction has been shown.

Lesion Limitation.

Palzer - *Protea cynaroides* controls infection by chemical factor. Phytoalexin production by hosts.

Marks - Kino formation as a measure of response to infection. Formation of kino spasmodic, due to change in interaction or ?

Weste - Temperature.

Marks - The amount of wood produced for the kino to form would more likely be moisture affected.

Podger - Kino formation not common, but lesion limitation is more common without kino formation.

Palzer - Why does the fungus stop growing.

Hinch - Using blue lupins, infected cells were high in gums. Lupins infected with P.c. high in hydroxy prolines; possibly strengthening the cell walls

Podger - Infections are compartmentalised (Shigo).

Disease Expression.

Palzer - Reduction in productivity. Foliage response to infection. Surface roots of jarrah subject to infection while deeper roots are not. Peter Kimber said jarrah had plenty of roots in water table. What is precipitating death in jarrah? Phytosynthesis, water loss, slow decline due to lack of nutrition. Temperature relation to water stress, stomatal closure increases leaf temperature by 15°C, antagonistic to enzymes.

Davison - Relative infrequency of isolation of *P.c.* from jarrah roots. Can you say that the amount of roots required to be killed to kill trees? Fungus present but can't be isolated. Isolation methods, time of year, intensive sampling, effects of environment on nutrients, other organisms, insect pests - Ros Hart's toxin.

Compartmentalisation by jarrah, qualitative and quantitative.

Podger - It seems Dr. Davison suggests that *P.cinnamomi* is not the major cause of jarrah dieback.

Davison - I think you only produced patch dieback.

Podger - No, I introduced the pathogen and reproduced the disease and recovered the pathogen from the soil.

Palzer - Koch's postulates probably not essential.

Heather - I agree.

Gerretson-Cornell - Phytophthora can be primary or secondary cause.

Brown - Changes in pathogen in disease over time.

Podger - Dr. Davison has challenged the sacred cow.

Shea - Two diseases in W.A. forest - 1, Banksia, 2, jarrah. It is difficult to understand why jarrah dies after 30-40 years. The pathogen has gone, by the time disease expression appears.
Difficulty of sampling crown over time.
The affect in the crown too remote to site of infection.

Dell - Difference in water potential signifies disease expression.

Gerretson-Cornell - -

Hingston - Not much known. Leaves of jarrah have been analysed from "diseased" and healthy trees, with no difference being found. Amount of nutrients and fluctuation of nutrients not known. Leaves probably supplied at standard rate but fewer and smaller leaves.

Weste - Activated protons used in nutritional studies - iron, phosphorous, calcium, differences in diseased and healthy plants.

Havel - To Dr. Davison - Jarrah dieback - jarrah takes a long time to die due to degradation of the crown.

Davison - How much root necrosis needs to occur before disease expression in the crown in jarrah.

Marks - Exudates produced by Eucalypts complicate isolation due to complex polyphenol which are highly fungi-toxic.

Davison - I am not suggesting that *P.c.* is not there, but it is simplistic to say it is the only cause of jarrah dieback.

Podger - I am not saying *P.c.* is the only cause of jarrah dieback, but is the cause of the components of the jarrah forest, with jarrah dying later.

Davison - -

Podger - Jarrah crown deterioration.

- Old - Could it perhaps be that P.c. by killing off the susceptible spp. of the understorey, then jarrah with a grassy/sedge understorey which is adverse to jarrah and it declines.
- Roth - I am completely confused. I am impressed by the recuperative capacity of jarrah, and my impression is that we are getting root damage and the reduction of the crown is a response to the reduction of the roots.
- Gerretson-Cornell - Erratic pathogenicity in different Eucalypt spp. at different times or environmental conditions.
- Shea - The jarrah system, unlike agriculture system, which thrive on very poor soils to produce a timber tree, is living in a fine balance with its environment. Application of fertiliser increases disease.
- Marks
- Weste - This is an agricultural phenomenon.
- Palzer - By applying fertiliser, a chain of events occur to all the components of the soil and biosphere.
- Havel - 1500kg/hectare application of P to jarrah forest was fixed.
- Shea - Management might think fertiliser would be the answer.
- Hingston - Fertiliser could increase the disease when applied to the infection area, but applied to other areas, might be beneficial.
- Heather - In rehabilitation in lands in N.S.W., the trees that are fertilised are less drought resistant.
The Eucalypts have the capacity to recycle nutrients in their boles; even manage to survive in hostile environments by this phenomenon.
- Shea - This would support slow death in jarrah.
- Podger - Supports Lew Roth's theory with Bill Heather's evidence. Jarrah is not a big tree, but it has a small crown and small crown relative to its bole. Under sampling of jarrah roots conceded.
- Davison - I agree and more work on jarrah isolations should be made.

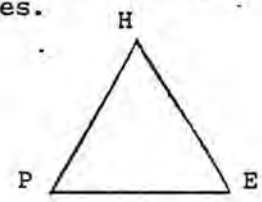
Epidemiology Analyses.

Procedures are now available

Shea - Measuring the disease has improved, especially with aerial photography, and I agree with your comments on epidemiological analyses.

12.35. Lunch.

13.35. Options for Control - W. Heather.



Apologies from Dr. Griffin, who is acting Vice Chancellor.

Disease is a host/pathogen reaction.

Research is largely confined to host and/or pathogen to the exclusion of the environmental factors.

The more factors you put into a system the more actions and interactions will emerge from the data. This may be difficult to analyse in management decisions and will rest on interpretation of the results.

Researchers tend to extrapolate from simplistic experimentation without the necessary inputs to the research.

Sufficient controls necessary to show up differences.

Forestry a large area land use with low return in income.

Two peculiarities of forest pathology compared to annual crops, costs will be

compounded for the length of the rotation, time for applying the control should be as early as possible for maximum return. It is necessary to have meaningful forms of control to assure time/money spent on a method of control will be economically feasible.

Allocation of research funds should have a potential of solving the problem.

Will managers comment on possible methods of control, bearing in mind practicability

1) Should we try and control jarrah dieback?

2) To prime jarrah forest only, or the whole forest?

Havel - Planting of dieback areas would lower yield of water, therefore allow natural regeneration.

Hopkins - Yes, we should control jarrah dieback.

Havel - Catchment protection in the Northern jarrah forest. Eastern jarrah forest more important for timber production.

Mulcahy - Priorities have been cost/benefit analysed in allocation of Research Grant.

Grace - Of the 4 metropolitan catchments, at 20¢ per kilolitre, the value of water alone is \$168 per hectare, per annum.

Podger - Should it be replaced.

Shea - We don't know what the effect would be removing jarrah, and whether replacement species will contain a salinity problem, which is vital to Perth water supplies.

Heather - If we do nothing, what would happen.

Shea - The result would be a chapparel, fynbos situation. The forest will not collapse in the next 10 years, but as the Banksia go out, the disease will be established and jarrah doomed.

Roth - I wish to support Dr. Shea's recent comments. I concur with most of what Bill Heather said but does that help the biologist.

Marks - I have tried to mitigate disease.

Roth - I believe our track record is miserable. Research on Port Orford Cedar - logs were selling at \$2 per 1,000. Today they are worth \$1,000 per 1,000.
Land value 20 years ago - \$3,500.
" " now - \$12,500.

These are management decisions and not biologist decisions.

The Dothistroma control method in N.Z. was a fine example of control of a disease at low cost with maximum effect.

Considerations of management should not inhibit biologist research.

Heather - Fire as an option of control.

Grace - At the moment prescribed burning of 600,000 hectares by aerial ignition per annum, with local burns in relation to bauxite mining. A burning cycle of 5 to 10 years is the present practice, but burning at longer intervals, up to 12 years.

Shea - I think we can improve the forest by the regeneration of Acacia, which will benefit jarrah in the long term. When do you make a decision on what to do

Hopkins - Fire management - hot fires for fauna management - fire with legumes could be effective in parts of the forest, but whether fire breaks should be artificially produced and the areas burnt to clear the area.

Podger - Have you got places where you can have a big fire in places which would not be socially unacceptable.

Grace - We are getting photographic evidence that we could do this against P.c. Fire control at present of paramount importance socially.

Heather - Can someone explain this photographic interpretation.

Bob Chandler

- Deaths are not necessarily associated with dieback, necessity for reliable indicator species for dieback identification. A suite of examples of sites where disease expression can be detected for Dr. Shea to follow up and for photographic interpretation and likelihood for disease extension. It seems there are differences especially from west to east. Detection of the disease both photographically and in isolation methods needs to be improved.

Prof. O'Connor

Murdoch - ? not represented.

Hygiene, Quarantine.

Roger

Underwood - Immediate prospect of control is dieback hygiene and quarantine, but there are still areas where disease or lack of it has not been established. Three quarters of the jarrah forest now under quarantine. Quarantine has been in practice for more than 3 years in the Northern jarrah forest and 3 in the Southern jarrah forest, and has been highly effective on the major spreading vectors, heavy machinery, etc., though not for trail bikes, etc.

Split phase logging, road stabilisation and.....important steps undertaken.

Fire control option now can be 'do nothing' rather than spread disease by fire control.

Podger - Detection. Hardy perennial that techniques are not sensitive but maybe sampling is not adequate.

Shea - 66% of samples proved positive from samples supplied by Chandler's group.

Hopkins - 80-90% result expected.

Shea - We get 90% from known dieback areas, i.e. Banksia sites.

Underwood - 32% in dieback gullies. Largely a sampling problem. Where should samples be taken?

Fertilising.

Heather - Nick Malaczuk - you and your group have a large fertiliser experiment in the jarrah forest.

Hingston - Yes, we are about to embark on a fertiliser experiment. A great deal of work has gone into preliminary monitoring and trial tests into N and P response will be started.

Grace - Are levels economic.

Hingston - Low level cheap, higher level 3 or 4 times the low. Low level may change understorey. We hope the higher level will help jarrah.

Warcup - Have you used the fertilisers to eliminate *Banksia* sp.

Hingston - This could be possible in the phosphate applications.

Warcup -

Shea - Is there possibility that species could be intolerant to high levels of P.

Heather - Yes, in snowgum, following heavy litter fall, the..... is sometimes accumulated in the wood.

Podger - Allan Hatch's work on P 20 years ago, no response from 20cwt P application.

Hopkins - He did not get growth response.

Hingston - Was it because he did not include nitrogen.

Hopkins - Yes, this is what he found.

Havel - Confirmed.

Marks - 1972 - fertiliser experiments in plantations startling response which required repeating after 2 years.

Fertilisers more important than water in Victoria.

Grace - Replacement species trials valuable for rehabilitation of dieback areas.

Heather - I agree; for example *Alnus*, which improves the soil conditions

Old - Calcareous amendments in W.A. Any progress?

Shea - This will be 3rd stop tomorrow - Jan, can you comment?

Titze - Nothing much to report. Pot trials with Dolomite lime 30% *Banksia* remained healthy. Field trials have not shown control of spread of the fungus.

Fungicides.

Hopkins - Sterilising soils for hygiene practice.

John ? - Field trial not yet providing results.

? - What rates.

John ? - 0.5g $\square^m$ a.i. - 2.5g $\square^{2m}$ a.i.

Podger - Treatment of landing sites must ensure all roots are reached.

Grace - What product should be used as fungicides in landing areas. Copper sulphate superseded by Sodium hypochlorite.
Bauxite landing sites.

Kagi - Sodium hypochlorite pH sensitive in sterilising effect.

Breeding for resistance.

Heather - I think breeding in perennials not an option, due to cultivar interactions.

Marks - 62 different lines of mixed spp. of eucalypt, resistant to P.c. Collect seed and trial of seedlings in progress.

A long term trial, but worth trying.

Heather - I agree selection for resistance is worth while.

13.40. Summing Up.

Roth - This meeting has raised an awful lot of questions. I wonder how many answers.

I am sure it has been stimulating to the researchers, but I hope not disappointing for the managers.

I think that after review of the results over the past 10 years, there should be a further meeting of active workers in the field.

Do not be constrained in your research by economics, but research must be as relevant as possible in an emergency situation, which is prevailing in W.A.

Could I suggest this notion -

Maybe each of us keep before us a picture of the system of the disease, and peripheral research should be funded by some other than emergency funds.

A single panacea is unlikely and that it will be a combination of efforts. Each of the many facets of the ecosystem will have to be dealt with.

Frank Podger commented we should go back and read our papers, and I agree; can we not conclude methods of control of inoculum, systems to cool the soil, can we toxify soil at optimum times. We must always keep in mind time, and aim our efforts to these crucial periods.

.....

Areas of needed research - a couple of pages.

Emphasis placed on methodology, especially in field situation compared to the laboratory, to be commended.

In general, overall research program is splendid and thanks for inviting me.

Joe Havel - General Discussion and Conclusions.

13.55. Research/management interface.

Observation → theory or hypothesis → testing → reformulate the theory → field testing (testing of feasibility).

Report → testing of the economic feasibility → social feasibility → and all its implications → legislation → funding → implementation.

Quarantine has probably been through these steps.

(When Frank Podger identified *P.c.* as the cause of dieback, he overstated the case and resulted in no further work starting for several years - he then followed through the steps.)

Marks - Steps need to be retraced after implementation.

Weste - W.A. Forest Dept. congratulated on obtaining quarantine.

Havel - Feasibility of spread by animals was not considered before quarantine, which has resulted in an explosion of the pig population due to exclusion of hunters.

Roth - Work at Dwellingup commended.

Havel - A force of necessity due to pressure groups, but there is a limit to this where a theory is half baked.

The media is inclined to feed on conflict.

Podger - Concur with congratulations, but Tasmania and N.S.W. also have achieved success in this way.

Havel - 16.20 Close.