

Gnangara Lake Vegetation Disease Survey

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GLOSSARY

Armillaria luteobubalina

A native wood rotting fungus that attacks the sapwood of living plants.

Autonomous disease spread

The natural movement of infective motile spores in saturated soils and growth of mycelium between connecting root systems.

Cambium

The layer of conductive tissue between the wood and outer bark of vascular plants.

Chlamydospore

A thick walled spore approximately 0.04 mm in diameter produced vegetatively by the rounding up of cells. Can tolerate drier conditions than zoospore.

Cryptic disease

The situation in which a host plant is infected with a pathogen but there are no observable secondary symptoms.

Dieback

In Western Australia, the term is specifically used to describe the disease in native vegetation caused by the *Phytophthora cinnamomi* fungus. The term is also generally used to describe any progressive deterioration of tree crowns.

Disease

The decline in plant health as a result of the presence of a pathogen and environmental conditions favourable to the pathogen.

Impact

The effect of disease on plant health.

Inoculum

Infective propagules capable of causing disease.

Keystone species

A plant species in an ecosystem that provides conditions pivotal for the survival of other flora and fauna.

Mycelium

A mass of fungal hyphae forming the body of a fungus.

Non-susceptible species

Plant species that do not display secondary symptoms of *Phytophthora cinnamomi* infection when exposed to the pathogen. They may display primary

symptoms of infection including the death and discolouration of root tissue and lesion development.

Phytophthora

A genus of microscopic fungi responsible for widespread damage in native vegetation of Western Australia.

Sporangium

An oval or ellipsoid spore sac approximately 0.057 x 0.033 mm that produces zoospores within a cell wall.

Susceptible species

Plant species that will develop secondary symptoms of *Phytophthora cinnamomi* infection when exposed to the pathogen. Secondary symptoms include crown decline and death of the host.

Vectored inoculum spread

The spread of infective propagules through the movement of infected soil or vegetative material. Agents include vehicles and machinery.

Vegetation associations

A group of similar plants that grow in a uniform environment that contains one or more dominant species.

Zoospore

A motile spore approximately 0.01 mm in diameter produced within a sporangium.

1. BACKGROUND

The plant disease known in Western Australia as "dieback" or "Jarrah dieback" is caused by an introduced, microscopic, soil-borne fungus of the genus *Phytophthora*. The most destructive and widespread species is *Phytophthora cinnamomi* Rands, which has caused irreversible decline of susceptible species from Eneabba in the north to Cape Arid on the south coast (Shearer & Tippett, 1989).

The *P. cinnamomi* fungus is highly invasive and will infect the roots of a large range of plant species. The primary symptom of infection is the death and discolouration of root tissue. The secondary symptoms are crown decline and/or the death of the infected plant. Plant species that are partially resistant and do not develop secondary symptoms are classified as "non-susceptible" hosts, and species that develop secondary symptoms are regarded as "susceptible" hosts. There are few native plant species that have been found to be completely resistant to dieback and able to inhibit fungal growth at the point of entry.

P. cinnamomi mycelial strands spread through the root system of an infected plant and can infect new hosts through direct root contact. Under certain environmental conditions, the mycelium develops sporangia. The sporangia release zoospores into the soil where they spread through water movement and, to a lesser degree, through self-propulsion. People can also spread the spores through the movement of infected soil or plant material.

P. cinnamomi has been inadvertently spread throughout the coastal sand plains and infected many areas. The impact that *P. cinnamomi* will have in native vegetation communities is determined by a complex interaction between the host species, the *P. cinnamomi* fungus and environmental factors. The Banksia woodlands of the Swan Coastal Plain are especially at risk as they contain many susceptible species.

Until recently, the only method of disease control was to implement hygiene measures designed to minimise vectored inoculum spread. There was no effective method of controlling autonomous disease spread within infected areas, or protecting susceptible plant communities in high conservation value areas. Several trials undertaken by the Department of Conservation and Land Management have shown that foliar spraying and stem injection of phosphorous acid gives excellent control of *P. cinnamomi* in a range of susceptible hosts. In a report to the Western Australian Minister for the Environment, phosphorous acid treatment is identified as an effective measure for the protection of threatened flora (Podger, James & Mulcahy, 1996).

Armillaria luteobubalina is a native fungus belonging to the basidiomycetes group of wood rotting fungi. The fungus is readily recognisable by characteristic white mycelial fans in the cambium of infected plants, and the presence of fruiting bodies (basidiomes) in June and July. Infection occurs through aerial dispersal of basidiospores released from basidiomes or through root to root transmission of mycelium, which results in discontinuous and discrete distribution of infections.

The fungus is a primary pathogen and has a wide range of host species. There are many hosts that are not susceptible to *Phytophthora cinnamomi* that are susceptible to *Armillaria luteobubalina* infection. The impact that an infection will have in a vegetation community depends on many factors including density of susceptible species, stress levels, site, soil, and climatic conditions. The disease management strategy for *Armillaria* is to segregate infested areas from uninfested areas during operations involving soil movement.

2. INTRODUCTION

This survey was commissioned by the Department of Conservation and Land Management as part of the environmental survey of Gnangara Lake and surrounding native vegetation with the following objectives:

- To determine the distribution of disease caused by the plant pathogen *P. cinnamomi* within native vegetation and demarcate any active disease fronts.
- Collect soil and root tissue samples for laboratory analysis to confirm the disease status and identify any species of *Phytophthora*.
- Determine potential hygiene risks associated with human activities within the survey area.
- Formulate strategies to minimise the potential for disease spread.

Field assessment commenced on 8 July 2001 and was completed by 11 July 2001. The method of disease assessment was the systematic examination of susceptible species for secondary symptoms of *P. cinnamomi* infection, supported by soil and root sampling. Assessment was also made for the predicted impact of *P. cinnamomi* in vegetation associations, disease distribution and impact, site disturbance and potential inoculum vectors.

3. METHOD OF DISEASE SURVEY

3.1 Field Assessment

The survey was conducted in two parts. An initial assessment was made from a slow moving vehicle traversing all open tracks, followed by walking transects with a 50 metre interval through areas with no vehicle access. The disease status was determined by observations of the health and presence of susceptible species.

Where secondary symptoms indicative of *P. cinnamomi* infestation were detected, the site was assessed for chronological disease pattern development and evidence of agents other than *P. cinnamomi* (such as fire, drought, lightning, other fungi and mechanical damage). Sites within the survey area that could not be assessed due to a low representation or absence of susceptible species were classified as uninterpretable.

3.2 Classification of Vegetation

During the disease survey, assessment was made of the vegetation structure and its susceptibility to *P. cinnamomi*. The vegetation was classified into vegetation associations based on the presence of one or more dominant species and the relative abundance of *P. cinnamomi* susceptible species was noted. The response that individual susceptible species will have to *P. cinnamomi* can vary and is dependent upon a complex relationship between the environment, the pathogen and the host. Observations of disease impact within existing infestations were assessed to predict the impact *P. cinnamomi* may have if introduced to uninfested vegetation associations of similar structure.

3.3 Sample Recovery

For each site where there were secondary symptoms and chronological disease pattern development consistent with *P. cinnamomi* infestation, a soil and root tissue sample was recovered for laboratory analysis. It is not possible to conclusively identify *P. cinnamomi* in the field as the fungus is microscopic during all stages of its life cycle. The sample sites were demarcated in the field with dayglow orange flagging tape and an aluminium identification tag displaying the sample identification number and date.

The samples were processed by the Conservation and Land Management Vegetation Health Service laboratory. The sample material was divided evenly into two containers and flooded with distilled water containing 15 *Eucalyptus sieberi* cotyledons that had been propagated in sterile conditions. The containers were covered and incubated at 25⁰ Celsius for ten days. Cotyledons that

changed colour from purple/green to brown were plated on antibiotic agar plates selective for *Phytophthora*. Plated material was left for five days then examined for the development of fungi. Identification of *Phytophthora* species was determined by the structure, size and development of sporangia and hyphae.

3.4 Mapping and Demarcation

A Department of Land Administration 1:20 000 colour aerial photograph from the Metro/Regional series (05/02/2000) covering the survey area was obtained from the Department of Conservation and Land Management as a Jpeg image. The photograph was used to identify the survey boundaries, internal tracks and possible infestations.

During the survey, areas of native vegetation displaying secondary symptoms and chronological pattern development consistent with *P. cinnamomi* infestation and classified as infested were plotted directly onto a print of the photograph. Segments of vegetation with too few susceptible indicator species and areas cleared of vegetation were classified as uninterpretable and were also plotted.

Active disease fronts were demarcated with 25 millimetre dayglow orange flagging tape tied to vegetation approximately 20 metres from the last secondary symptoms. The resulting demarcation boundary is parallel to the disease front within uninfested vegetation, with a twenty metre buffer to allow for cryptic infestations and autonomous disease spread for several years. The demarcation boundary was plotted directly onto a print of the photograph.

The plotted boundaries were transferred from the photograph to the Jpeg image provided by the Department of Conservation and Land Management Forest Management Branch. This information forms the "*Phytophthora cinnamomi* Hygiene Management Map" presented in Appendix 1.

4. RESULTS

4.1 Vegetation Associations

The native vegetation within the survey area was classified into three broad vegetation associations based on the presence of one or more dominant overstorey species. The vegetation associations and their susceptibility to *P. cinnamomi* infestation and spatial distribution are listed below.

Banksia Woodland

This vegetation association consists of an open woodland of *Banksia attenuata*

and *Banksia menziesii* with occasional *Banksia illicifolia* and *Nuytsia floribunda* over low heath including *A. cygnorum*, *Hibbertia* spp., *Patersonia* spp., *Stirlingia latifolia* and *Xanthorrhoea preissii*. The majority of the flora within the Banksia woodland is susceptible to *P. cinnamomi* and infested sites were displaying extremely high impact (90% or higher mortality) in the Banksia overstorey and moderate to very high impact (30% to 90% mortality) in the understorey.

There is a large segment of Banksia Woodland on a sandy rise adjacent to and east of Lake Gnangara comprising approximately one quarter of the survey area.

Fringing Melaleuca Wetland

This vegetation association consists of a woodland of *Melaleuca raphiophylla* (Paperbark) and occasional *Eucalyptus rudis* with a mix of understorey species including *A. cygnorum*, *Melaleuca* spp. and sedges. The predicted impact of *P. cinnamomi* is negligible as there is a very low representation of susceptible species and the wetland was classified as uninterpretable.

The fringing Melaleuca wetland surrounds Lake Gnangara above the high water mark and extends south to Gnangara Road and west to the assessment boundary. The vegetation association includes transitional vegetation zones including open sedges and *Corymbia calophylla* (Marri) woodland on the eastern lake perimeter.

Melaleuca Woodland

This vegetation association consists of an open woodland of Paperbark and occasional *Eucalyptus marginata* (Jarrah) and Marri over low heath of *A. cygnorum*, *Melaleuca* spp., *Scholtzia* spp. and *X. preissii*. Approximately 95% of the open Melaleuca woodland is infested with *P. cinnamomi* and is displaying low impact.

The Melaleuca woodland is to the east of the Banksia woodland and adjacent to the eastern survey boundary.

4.2 Disease Distribution

There is an infestation within the Melaleuca woodland on the eastern survey boundary that occupies approximately 20% of the survey area. Sample 2 was recovered from *B. attenuata* displaying secondary symptoms of infection and returned a positive result for *P. cinnamomi*. Active disease fronts were readily observable from secondary symptoms within the Banksia woodland immediately adjacent to the Melaleuca woodland.

There is an infestation within Banksia woodland on the eastern survey boundary that is circular and approximately 90 metres in diameter. Sample 3 was recovered from *B. attenuata* displaying secondary symptoms of infection and

returned a positive result for *P. cinnamomi*. The active disease front was readily observable from secondary symptoms in the Banksia overstorey and susceptible understorey species.

There is an infestation within Banksia woodland on the north eastern perimeter of Lake Gnangara that extends from the northern survey boundary to the main car park/amenities building. Sample 1 was recovered from *B. attenuata* displaying secondary symptoms of infection and returned a negative result. Positive identification of *A. luteobubalina* was made at the site. Active disease fronts were readily observable from secondary symptoms within the Banksia woodland.

There is relatively small infestation within Banksia woodland on the northern survey boundary adjacent to private property that is semi circular and approximately 20 metres in diameter. There were no suitable (recent) deaths in susceptible species to sample. There is evidence of chronological disease spread and deaths in susceptible species within remnant native vegetation on private property.

Positive field identification of *A. luteobubalina* was made within all three infestations through excavation of mycelium within root cambium and the presence of basidiomes.

4.3 Uninterpretable Areas

Lake Gnangara and the fringing Melaleuca wetland vegetation association were classified as uninterpretable due to the inadequate representation of susceptible species. The fringing Melaleuca wetland between the infestation to the north east of Lake Gnangara and the high water mark was classified as infested due to the high probability of cryptic infection.

A sand pit to the east of the car park/amenities building was classified as uninterpretable due to the absence of vegetation.

4.4 Field Demarcation

The western disease front of the infestation within the Melaleuca woodland was demarcated from the northern survey boundary to the southern survey boundary. No demarcation was made on the eastern boundary as the uninfested areas were considered too small to segregate. The two other infestations were demarcated. The uninterpretable areas were not demarcated.

5. DISCUSSION

The principle aims of *Phytophthora* management are to minimise the risk of increasing the occurrence of disease through vectored inoculum spread, and to minimise the impact of existing infections (Shearer and Tippett, 1989). The following discussion illustrates the issues that have been considered in analysing these risks and the potential impact of vectored and autonomous spread of disease within the survey area.

5.1 *Armillaria Luteobubalina* and *Phytophthora Cinnamomi*

From positive sample recovery of *P. cinnamomi* and positive identification of *A. luteobubalina* within the survey area, it is probable that the observable secondary symptoms may be attributable to one or both of the pathogens. Some species that are not susceptible to *P. cinnamomi* are susceptible to *A. luteobubalina* (such as Marri), while many are susceptible to both.

The negative sample return for *P. cinnamomi* at Sample Site 1 may be attributable to recovering the sample from *B. attenuata* cryptically infected with *A. luteobubalina* (sections of roots containing mycelium not excavated) and not *P. cinnamomi*. The active disease front was indicative of both pathogens being present, with some segments being continuous and discrete (indicative of *P. cinnamomi*) and others being discontinuous (indicative of *A. luteobubalina*).

5.2 Time and Mode Inoculum Vectoring

From the initial point of inoculum introduction, *P. cinnamomi* propagates through mycelium spread in susceptible species roots and under certain climatic conditions will produce motile spores (Zoospores). The relatively uniform distribution of Banksias and vertical drainage within the sandy soils results in a continuous disease front with consistent rates of spread in all directions. Infestations that are circular indicate inoculum was introduced at a central point and the disease has propagated equally in all directions. Infestations that are oval or rectangular indicate that there has been multiple points of inoculum introduction along a transect.

There is a significant range of rates of mycelium spread in Banksia roots from as low as 20 centimetres per annum to several metres per annum (Shea & Dillon, 1980). The rate of autonomous disease spread is dependent on soil type, geology, vegetation structure, soil moisture and temperature. It is possible that the rate of spread may have varied in the past with fluctuations in soil moisture and temperature. Based on experience and field observations of the decay of Banksia stags (dead trees) and their position in relation to the active disease

front, it is probable that the rate of autonomous disease spread in Banksia woodland has been relatively constant at approximately one metre per annum.

It was possible to make estimations of the time and mode of inoculum vectoring based on the size and shape of infestations. The infestation in the Melaleuca woodland on the eastern survey boundary has been present for many decades and it is probable that susceptible species that may have been present (particularly Banksias) have been decimated. The relative expanse, convoluted disease boundary and large isolated infestation at Sample Site 3 suggest multiple points of inoculum introduction. There are many old excavation sites (possibly exploration for peat) that appear to be associated with pathogen vectoring within the survey area, and possibly the initial introduction of the pathogen.

The size and shape of the infestation on the north eastern perimeter of Lake Gnangara suggests multiple points of inoculum vectoring along a transect. There is a vehicle track within the infestation parallel to the Gnangara Lake high water mark that is the probable pathogen vector. The distance between the active disease front to the east of the track and the track varies from 20 metres to 50 metres, indicating multiple points of inoculum vectoring as long as 50 years ago. It is possible that inoculum was vectored during establishment of the track through the importation of infected soil.

The infestation adjacent to private property on the northern survey boundary is semi circular (adjacent to a cleared track/fire break) and approximately 20 metres in diameter, suggesting a point source of inoculum approximately 10 years ago. The inoculum may have been introduced during establishment, maintenance or use of the adjacent track, or autonomously spread from adjacent private property where there are secondary symptoms indicative of infestation.

5.3 Potential Inoculum Vectors

Any activity that has the potential to import infected soil or vegetative material to uninfested areas has the potential to initiate new infestations. The probability of initiating new infestations is dependant upon many factors including the type (mycelium, zoospores, chlamydospores) and volume of propagule vectored, the soil type/moisture content and the proximity of hosts.

Activities that involve movement of large quantities of soil (such as road building, mining) within susceptible vegetation are considered high risk. Activities that involve small quantities of soil movement (such as bush walking) are considered low risk. Evidence suggests that the infestations within the survey area were established through relatively high risk activities in the past. The absence of small/new infestations within susceptible vegetation suggests there has been no recent viable inoculum vectoring. The only current/recent activity identified

during the survey with the potential for inoculum was extensive use of vehicle tracks by off road vehicles.

As there is regular vehicle access without hygiene constraint, it is probable that there is soil movement during moist soil conditions. The reasons for inoculum not being viably vectored to uninfested areas are beyond the scope of this report, but may include low levels of inoculum in infested soils, the type of inoculum present, low levels of infested soil movement, no survival of vectored inoculum, or a combination of these factors.

5.4 Predicted Impact of Autonomous Disease Spread

The Banksia woodland vegetation association to the east of Lake Gnangara is extremely susceptible to *P. cinnamomi*. Infestations are displaying extremely high impact, with total loss of the Banksia overstorey and moderate to very high impact in the understorey. Many Banksias are keystone species (Shearer, Wills & Stukely, 1991), providing a food source for insects, birds and mammals. The loss of these keystone species will have a negative impact on the populations of dependent animal species as well as the aesthetic and intrinsic values of the native plant community.

P. cinnamomi is spreading autonomously in the Banksia woodland at a rate of approximately one metre per year. With active disease fronts along the western and eastern extents, the Banksia woodland will become completely infested over time. The smaller areas of uninfested Banksia woodlands on the eastern boundary will be completely infested within 30 to 40 years. The autonomous spread of *P. cinnamomi* can be reduced with the application of phosphorous acid (a cheap, biodegradable and non-toxic chemical), that has been found to be very effective as both a curative and protectant against *P. cinnamomi* in a range of susceptible hosts (Komorek, Shearer, Smith & Fairman, 1997).

5.5 Cryptic Infestation

It is probable that there is some degree of cryptic infestation within the fringing Melaleuca wetlands. The absence of susceptible species (particularly Banksia) may indicate that the areas have been infested for many years. It is not practical to determine disease distribution through random sampling. It is possible that small cryptic infestations may occur within the Banksia woodland that were not detected during the survey, due to an absence of secondary symptoms indicative of *P. cinnamomi* infestation. The probability of this is low due to the absence of recent high risk activities.

6. RECOMMENDATIONS FOR MANAGEMENT

It is recommended areas of uninfested Banksia woodland are protected from vectored pathogen spread. Any activities with a potential risk of moving infected soil or vegetative material to uninfested Banksia woodland should be analysed to determine the level of risk. Activities such as road/fire break construction and planting of nursery stock are considered high potential risk of viable inoculum vectoring and hygiene procedures should be established for any planned activities.

It is recommended that control of vehicle access is considered. Unrestricted vehicle access to the uninfested Banksia woodland does present a potential risk of inoculum vectoring, although evidence suggests that there is a low risk of viable inoculum vectoring (establishment of new infestations). Consideration should be given to the resource allocation required to achieve control of vehicle access in relation to the relatively low hygiene risk.

It is recommended that the viability of a strategic phosphorous acid application program to control autonomous disease spread is investigated. Methods of application that can be considered include aerial (aircraft) spraying, foliar spraying and stem injection.

It is recommended that only resistant plant species should be planted within uninterpretable or infested areas during revegetation projects. Appendix 2 lists some species that may be considered for rehabilitation planting. Appendix 3 lists susceptible species that should be excluded from rehabilitation planting.

7. REFERENCES

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