Department of Environment and Conservation

Vegetation Health Service

Annual Report 2009-2010

Phytophthora Detection

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1. Introduction

The **Vegetation Health Service (VHS)** provides a dedicated, specialist scientific service for the detection and identification of *Phytophthora* species from samples associated with the management of the State's forest and conservation estate, logging and mining activities, private industry, and research. The VHS is funded by DEC's Sustainable Forest Management Division (90%) and Nature Conservation Division (10%). It is run by Science Division staff based at DEC's Kensington Research Centre. Its services are offered free of charge to all DEC sections and personnel, and to non-profit community groups sponsored by DEC, such as SCRIPT and the NRM groups. The service is also available to external clients at a standard fee of $77 per sample, with discounted rates applying to samples collected for Alcoa World Alumina Australia, the Forest Products Commission (FPC), and Caring for our Country (formerly NHT) projects. No other *Phytophthora* testing service is available on this scale in WA.

> **Processing of samples**: Samples received by the VHS in most cases include a mixture of soil and plant-root material, which is baited for *Phytophthora* using the *Eucalyptus sieberi* cotyledon baiting method. Bait material is plated to selective agar medium for incubation, and any possible *Phytophthora* colonies that emerge are then isolated to pure culture for identification to species. Where isolation of the pathogen from specific host-plant tissue is required, roots are surface-sterilised and direct-plated to selective agar medium for *Phytophthora* isolation. All isolates are identified to species in the VHS from their morphological characters, and **DNA sequencing** is used for selected isolates through the service provided by the Centre for *Phytophthora* Science and Management (CPSM) at Murdoch University.

Results are supplied to clients as soon as possible. All results (both *Phytophthora*-positive and negative) are added to the **VHS database**, along with details of sampling location, associated plant species, land tenure, etc. The database now contains 34,951 records (at 30th June 2010), and this information resource is made available to land managers and researchers as required. Representative *Phytophthora* cultures are added to the **VHS Culture Collection** (see **Section 3** below). This now contains 995 cultures, plus earlier collections. Cultures from the Collection are made available to researchers in DEC and the CPSM, and in other institutions, on request.

The VHS also provides advice to assist Departmental staff, and also the public, with other plant disease problems in forests, plantations, parks and reserves, and nurseries.

2. Annual summary – samples processed

During the 2009-2010 financial year, the VHS received 1,581 samples for testing for the presence of *Phytophthora* (**Table 1** and **Figure 1**). This was a little below the previous year's total of 1,784 (see **Table 4** and **Figure 4**). The sources of these samples are as follows:

DEC – samples sent by **DEC Dieback Interpreters** (or by contractors to DEC), and **District staff**, in fulfilment of DEC's forest and land management responsibilities.

BCI – samples sent by DEC personnel associated with the **Biodiversity Conservation Initiative**.

ALCOA – samples sent directly by **Alcoa World Alumina Australia** (or by contractors to Alcoa) in fulfilment of Alcoa's forest management responsibilities prior to, during and after mining activities, on the DEC estate.

PRIVATE – samples sent directly by external clients (land managers or private contractors).

RECOUP – samples for external clients sent by DEC dieback interpreters.

FPC – samples sent directly by, or for, the **Forest Products Commission**, including FPC Nurseries.

CC – samples associated with projects funded by **Caring for our Country** (formerly **Natural Heritage Trust, NHT)**.

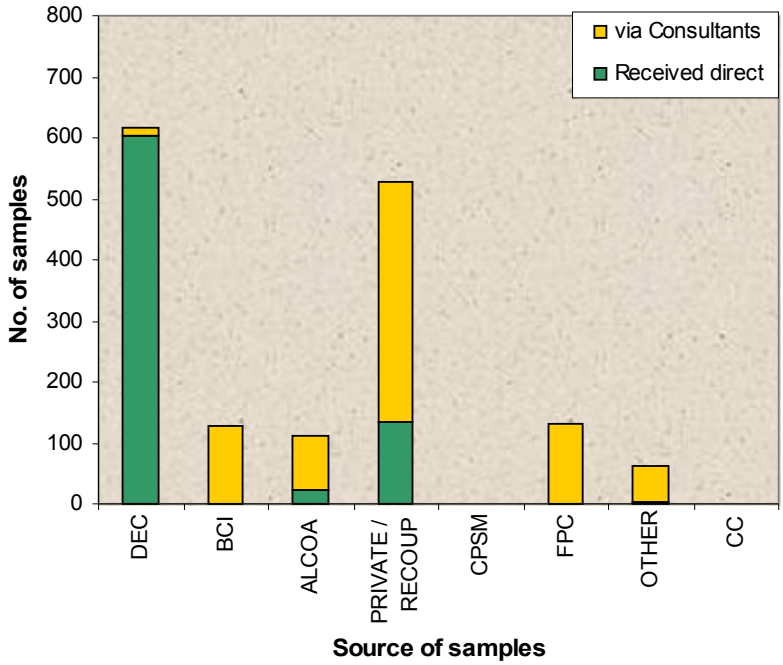
CPSM – samples associated with projects carried out at the **Centre for *Phytophthora* Science and Management** at Murdoch University.

The majority of samples processed by the VHS are collected by, or for, DEC. These include samples sent by FMB Dieback Interpreters from DEC-managed forest being monitored for *Phytophthora* in conjunction with logging by the FPC. Whilst all sampling of Forest Blocks due for logging by FPC was previously done by FMB Dieback Interpreters, some has been carried out again this year by consultants and it appears that this trend will continue. Samples are also received from various DEC District offices (**Table 2** and **Figure 2**). An increased number of Consultants – Glevan Consulting, Dieback Treatment Services, NPC Consulting, Moore Mapping, RPS Environmental, Coffey Environments, Cardno, Ficifolia Consulting, Enviroworks Consulting, and NRG Consulting – supplied 972 samples from various sources including DEC land and Alcoa (**Table 1** and **Figure 1**). In addition to Alcoa World Alumina Australia, mining companies sending samples direct to the VHS included Tiwest, Iluka Resources Ltd, Simcoa Operations, and Wesfarmers Premier Coal; samples were also received direct from Southern Gateway Alliance in association with the Perth-Bunbury Highway project. No samples were received from Caring for our Country (formerly NHT) projects in 2009-10. Other sources included NRM groups, government authorities, and shires.

VHS facilities have again been made available for the processing of samples for **other DEC research projects** (Dr Chris Dunne).

Table 1 and Figure 1. Numbers of samples received from major sources, including consultants, in 2009-2010. Numbers of samples received from the same sources in 2008-2009 are shown in the Table for comparison. Abbreviations of sources are as listed above.

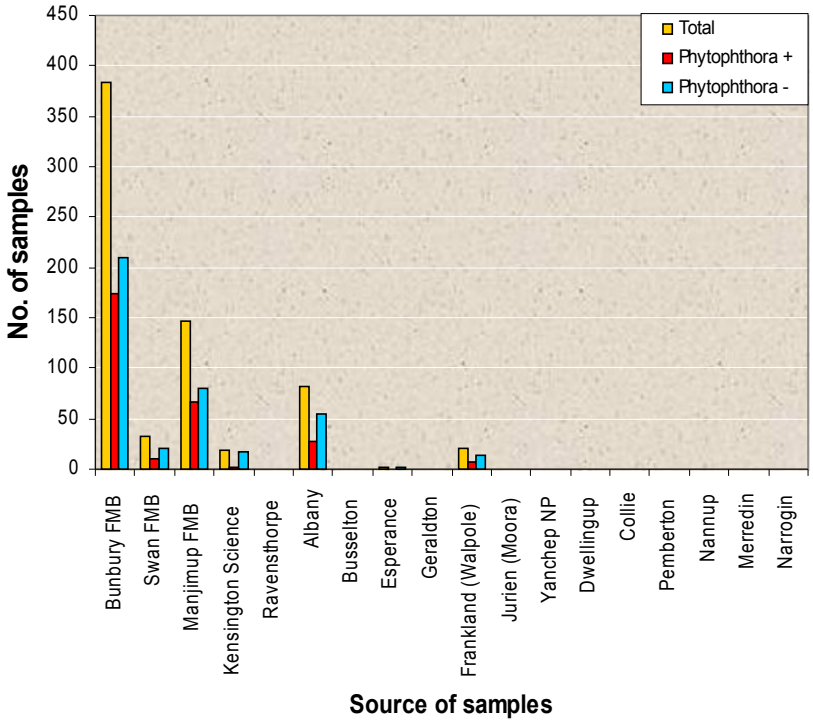
Source	No. of samples			
	Received direct	Received via Consultants	Total 2009-2010	Total 2008-2009
DEC	603	15	618	941
BCI	0	128	128	6
ALCOA	22	90	112	58
PRIVATE / RECOUP	133	396	529	510
CPSM	0	0	0	0
FPC	0	131	131	239
OTHER	3	60	63	30
CC	0	0	0	0
TOTAL	**761**	**820**	**1581**	**1784**


800
700
600
500
400
300
200
100
0
No. of samples
via Consultants
Received direct
DEC
BCI
ALCOA
PRIVATE / RECOUP
CPSM
FPC
OTHER
CC
Source of samples

Details of the sources (by District) of DEC samples received for the year are shown in **Table 2** and **Figure 2** (including Recoup samples, but excluding BCI samples and DEC samples collected by consultants). The locations where DEC and other samples were collected are shown on the attached **Map**, with results broken down to distinguish *P. cinnamomi*, Other *Phytophthora* spp. (including undescribed new *Phytophthora* taxa), and Negative.

Table 2 and Figure 2. Numbers of samples received from DEC offices in 2009-2010, and numbers giving positive or negative recoveries of *Phytophthora*. Total numbers of samples received from the same sources in 2008-2009 are shown in the Table for comparison. DEC samples received from consultants are not included.

DEC Office	No. of Samples			
	Total received 2009-2010	***Ph*. Positive 2009-2010**	***Ph*. Negative 2009-2010**	**Total received 2008-2009**
Bunbury FMB	384	174	210	451
Swan FMB	32	11	21	35
Manjimup FMB	146	66	80	94
Kensington Science	19	2	17	10
Ravensthorpe	0	0	0	2
Albany	81	27	54	208
Busselton	0	0	0	0
Esperance	2	0	2	0
Geraldton	0	0	0	0
Frankland (Walpole)	20	6	14	10
Jurien (Moora)	0	0	0	1
Yanchep NP	0	0	0	0
Dwellingup	0	0	0	0
Collie	0	0	0	0
Pemberton	0	0	0	2
Nannup	0	0	0	0
Merredin	0	0	0	0
Narrogin	0	0	0	0
TOTAL	**684**	**286**	**398**	**813**


Total
Phytophthora +
Phytophthora -
No. of samples
450
400
350
300
250
200
150
100
50
0
Bunbury FMB
Swan FMB
Manjimup FMB
Kensington Science
Ravensthorpe
Albany
Busselton
Esperance
Geraldton
Frankland (Walpole)
Jurien (Moora)
Yanchep NP
Dwellingup
Collie
Pemberton
Nannup
Merredin
Narrogin
Source of samples

3. *Phytophthora* species

Based upon the traditional methods of identification of *Phytophthora* isolates in pure culture by microscopic examination of their morphological characters, the following six species (in addition to *P. cinnamomi*) were previously believed to be present in WA natural ecosystems: ***P. citricola, P. megasperma, P. cryptogea, P. drechsleri, P. nicotianae*** (Stukely *et al*., 1997), and ***P. boehmeriae*** (D'Souza *et al*., 1997). Representative isolates of the WA species had been sent to CABI (UK) for verification.

In addition to the above species, ***P. inundata*** had been identified by DNA sequencing prior to June 2007 (Stukely et al., 2007b). Testing of new and historical isolates by this technique since 2005 has also led to the discovery of as many as **ten new and undescribed *Phytophthora* taxa**, designated "**P.spp.1–12"**, from dying native vegetation in WA (see **Section 3.1**) (Stukely et al., 2007c; Stukely et al., 2008; Burgess et al., 2009). The first of these new species ('Psp4') has been formally described as *Phytophthora multivora* sp. nov. (Scott et al., 2009).

During 2009-2010, *Phytophthora* was isolated from 507 of a total of 1,581 samples processed (compared with 711 of 1,784 in 2008-2009) (**Table 3, Figure 3**). ***Phytophthora cinnamomi*** was, as usual, the species most frequently isolated (443 samples). ***P. cryptogea*** was isolated from seven samples, ***P. inundata*** from two, and ***P. nicotianae*** from one.

The newly described species ***P. multivora***, closely related to *P. citricola* (and morphologically very similar to it), was isolated from 42 samples and identified by DNA sequencing. Sixteen samples gave isolates that were shown by DNA sequencing to be members of other new (as yet undescribed) *Phytophthora* taxa (see **Section 3.1**): **Psp 1** (three isolates), **Psp9** (one), **Psp7** (seven), **Psp3** (one), **Psp8** (one), and **unique isolates** (three).

Four samples yielded both *P. cinnamomi* and *P. multivora*.

The locations where DEC and other samples were collected are shown on the attached **Map**, with results categorised to distinguish a). *P. cinnamomi*, b). Other *Phytophthora* spp. (including the undescribed new *Phytophthora* taxa), and c). Negative.

Representative pure cultures of all *Phytophthora* species isolated, and cultures of individual species representing different geographic locations, ecosystems, or host plants, or morphological types, were added to the permanent **VHS Culture Collection**. These cultures are maintained in a pure and viable condition by periodically sub-culturing, checking their purity and establishing fresh storage cultures. There are now 990 cultures stored in the main VHS Collection. In addition, the VHS holds *Phytophthora* cultures from the following earlier collections that were stored and numbered by researchers and consultants: MJS series (M Stukely), DCE (E Davison), TCH (T Hill) and HSA (Hart, Simpson and Associates). Cultures from the Collection are made available to researchers within DEC and the CPSM, and in other institutions, on request. This culture collection is a unique, and very valuable, scientific resource.

DNA sequencing of historical isolates from the Collection has continued (see Section 3.1).

Table 3 and Figure 3. Numbers of samples from which *Phytophthora* species were isolated by the VHS in 2009-2010. Corresponding numbers of samples for 2008-2009 are shown in the Table for comparison.

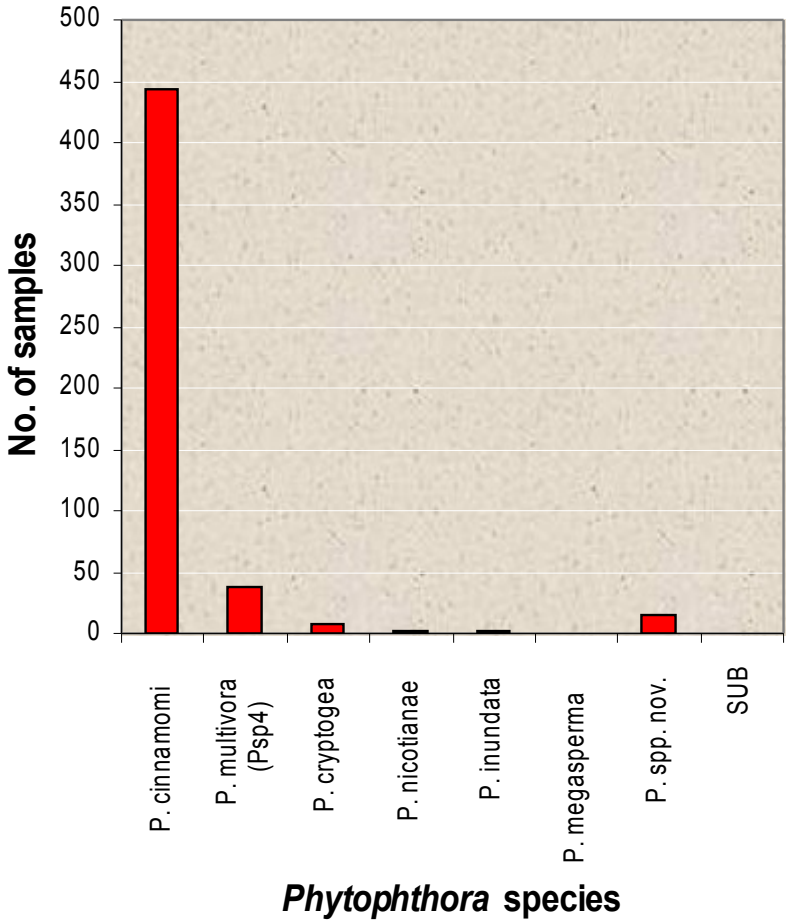
***Phytophthora* species**	**No. of samples with positive *Phytophthora* recovery 2009-2010**		**No. of samples with positive *Phytophthora* recovery 2008-2009**	
	Primary result	**Second species[1]**	**Primary result**	**Second species[1]**
P. cinnamomi	443		648	
P. multivora[2] *('Psp4')*	38	4	32	1
P. cryptogea	7		7	
P. nicotianae	1		9	
P. inundata	2		0	1
P. megasperma	0		0	
P. spp. nov.[3]	16		9	
SUB[4]	0		6	
TOTAL	**507**	**4**	**711**	**2**

[1]Second species – in addition to *P. cinnamomi*, these samples yielded a second *Phytophthora* species as shown.

[2]*P. multivora*, described in 2009, was formerly identified as *P. citricola.*

[3]*P.* spp. nov. – these isolates represent undescribed new *Phytophthora* taxa 'P.spp.1-12' (see Section 3.1), excluding *P. multivora* ('Psp4').

[4]SUB – these *Phytophthora* cultures are still in the process of being identified.


500
450
400
350
300
250
200
150
100
50
0
No. of samples
P. cinnamomi
P. multivora (Psp4)
P. cryptogea
P. nicotianae
P. inundata
P. megasperma
P. spp. nov.
SUB
Phytophthora species

3.1. New *Phytophthora* records, and undescribed *Phytophthora* taxa in WA

The adoption and use of DNA sequencing techniques for the identification of *Phytophthora* species in the last decade has caused something of a revolution in *Phytophthora* taxonomy, worldwide. The availability of these techniques in WA, through the **Centre for *Phytophthora* Science and Management** (CPSM) at Murdoch University, has enabled the routine testing of some WA isolates that could not be identified satisfactorily by traditional morphological examination. In addition, DNA sequencing has confirmed that the *P. cinnamomi* isolates have been correctly identified from their morphology.

> DNA sequencing and data analysis: DNA was extracted from pure cultures of *Phytophthora* grown on half-strength potato dextrose agar, and the **Internal Transcribed Spacer (ITS)** regions of the rDNA were amplified using primers ITS6 and ITS4. BLASTn searches of sequence data were conducted in **GenBank** to determine the most closely related *Phytophthora* spp. Sequences were then aligned and parsimony and distance analyses conducted in PAUP. **Phylogenetic trees** were constructed (see **Attachment**).

Very significant progress has been made again in the last year. We have now received ITS rDNA sequence data for a total of 375 recent and historical isolates of *Phytophthora* (to 30 June 2010), most of which were obtained from samples collected in natural ecosystems in WA. Sequences of representative isolates of all undescribed taxa and newly recorded species from WA were deposited on GenBank. [**GenBank** is a large, international, public domain database of DNA sequences that was created in 1988.]

In brief, our findings to date on the new *Phytophthora* taxa are:

- There are now known to be as many as **ten undescribed new species of *Phytophthora* present in natural ecosystems in WA** (designated "**P. spp. 1 - 12**").
- The WA Phytophthoras show a very high level of diversity, representing eight of the ten phylogenetic clades (groups) of *Phytophthora* that are known, worldwide. See the **phylogenetic tree** (**Attachment**); explanatory notes are given in **Section 9** (p. 11).
- The first of the new WA species, ***P. multivora*** sp. nov. ['Psp4'], was described in 2009.
- In addition to the above ten new species (for each of which there are multiple isolates), there are several **unique *Phytophthora* isolates** from natural ecosystems in WA.
- There is strong new evidence (2009-2010) that some of the unique WA *Phytophthora* isolates are **hybrids**. Further investigation of these *Phytophthora* hybrids is in progress.
- Only **two WA isolates** of the many identified earlier as ***P. megasperma*** (based on their morphology) had a 100% match to sequences for *P. megasperma* on GenBank. The others represent several diverse new taxa.
- **None** of the many WA isolates identified earlier as ***P. citricola*** (based on their morphology) had a 100% match to sequences for *P. citricola* on GenBank. These isolates all represent new taxa, mostly *P. multivora* and some 'Psp2' (the latter has a distinct morphology).
- **None** of the WA isolates identified earlier as ***P. drechsleri*** (on their morphology) had a 100% match to sequences for *P. drechsleri* on GenBank. These isolates all represent new taxa.
- Many of the ten new taxa, and all of the unique isolates, have so far been recorded **only in WA**. A few of the new taxa are a close match to undescribed isolates found overseas.
- All but one ('Psp8') of the ten undescribed *Phytophthora* species have been found **associated with dying plants** in native forest or heath-land in WA.
- Some undescribed *Phytophthora* species, and some more hybrids, have also been isolated from rivers and streams in the "Fishing for *Phytophthora*" project at CPSM.
- Several of the new *Phytophthora* taxa are associated with deaths of plants representing multiple families.
- The number of plant species associated with the new Phytophthoras is still increasing rapidly, as new isolates are found.

- Two isolates of each of ***P. asparagi*** and the undescribed ***P.* taxon 'niederhauserii'** were identified by DNA sequencing. These represent new records of these species from natural vegetation in WA. They were all isolated from samples collected in 2006-2007.
- An undescribed isolate collected near Busselton in 2005, designated 'Psp6' and lodged on GenBank, was found in 2008 to be a 100% match to a pathogen of tobacco in crops in the USA. It has been provisionally named there as *P.* taxon 'personii'.
- WA isolates previously identified as *P. gonapodyides* align with the undescribed ***P.* taxon 'PgChlamydo'**.
- The large group of isolates designated earlier as Psp5 are now included as ***P. cryptogea***.

3.2 Management implications – the need for further investigation of new taxa

Many of the undescribed new *Phytophthora* taxa (reported in **Section 3.1**) are genetically quite distinct from the other Phytophthoras found to date in natural ecosystems in WA, despite some of them having some strong physical similarities in culture to known *Phytophthora* species (eg *P. citricola, P. megasperma, P. drechsleri*). These genetic differences are cause for concern. They suggest that the 'new' Phytophthoras may have a different set of capabilities, strengths and vulnerabilities, and may pose different potential and immediate levels of threat to biodiversity, which must now be investigated.

It is imperative that these undescribed Phytophthoras are all properly described and documented as separate, individual components of our biodiversity.

The undescribed Phytophthoras, and the newly recorded ones, are all potential pathogens, as indicated by their associations with dying native flora that have already been recorded. It must be remembered that no *Phytophthora* has yet been documented as a "benign" organism – all are pathogens, with host specificities varying from a single plant species (eg *P. sojae*) to over 1,000 species (*P. cinnamomi*). Detailed investigations must therefore be conducted on each new *Phytophthora* species to determine its host range, pathogenicity, distribution, ease of spread, preferred environmental conditions, and other characteristics such as its response to the inhibitor, phosphite.

- It is not known whether these new *Phytophthora* taxa are **indigenous or introduced**.
- New evidence (2009-2010) shows that several unique *Phytophthora* **hybrids** are present.

Some of the new Phytophthoras (eg *P. multivora* ['Psp4'], formerly identified as '*P. citricola'*), but not all, are widespread and are associated mostly with low-impact dieback sites. **However, it is important at this stage that they should not all be treated as indigenous (and hence "not threatening"), until they have been investigated further.** It appears that they are not particularly new introductions, with some isolates of most of them having been obtained from samples collected in WA in the 1980s and 1990s, as well as more recently.

It is likely that the 'new' Phytophthoras will all have at least some level of pathogenic ability, and so it can be expected that they will damage native vegetation under some circumstances. It is known, for example, that the "*P. megasperma"* group causes most damage following summer rainfall in WA's coastal heath-land. Clearly, this high impact occurs at irregular intervals. It is also likely that increased levels of human activity in and around infested areas in the short term, together with the consequences of climate change and other stressors in the longer term, will exacerbate the effects of at least some of these Phytophthoras on natural ecosystems.

In land management, the Precautionary Principle should be applied: the undescribed Phytophthoras and hybrids should now all be regarded as a threat, and managed accordingly, until it is proven otherwise. Consistent, pro-active efforts should be made to prevent their spread to non-infested areas. Clearly, this issue now needs to be addressed by land managers with a well-considered, adaptive management approach. Current, proven approaches and procedures used to manage *P. cinnamomi* are appropriate at this stage for managing all soil-borne Phytophthoras that are present.

Research projects (**Section 6**) will provide answers to some of the key questions above. **An estimate of the level of threat that each *Phytophthora* poses to our biodiversity is required,** so decisions can then be made, based on sound science, as to whether additional, specific management strategies involving intervention are appropriate or necessary. Controls applied currently for *Phytophthora cinnamomi* (such as phosphite application, and restricting operations to "dry soil" conditions) may not always be appropriate, nor directly applicable without modification, for managing some potential 'new' *Phytophthora* species.

The newly discovered ***Phytophthora*** **hybrids** and their origins are being investigated further. Their presence reinforces the importance and urgency of preventing the spread of Phytophthoras wherever possible. **The opportunities for new hybrids to form must also be minimised**.

In general, a pro-active response with intervention at the earliest possible opportunity, to confine and possibly eradicate small *Phytophthora* infestations in priority areas, can be expected to pay very large dividends in future nature conservation efforts and could also benefit the commercial sector. This applies to all *Phytophthora* species.

4. Historical record of VHS operations and samples processed

Since the Vegetation Health Service laboratory was established at Kensington Research Centre in 1992 (initially as the Dieback Detection Service, DDS), a total of 28,276 samples have been processed for *Phytophthora* detection (to 30th June 2010) (**Table 4** and **Figure 4**). The 2009-2010 total (1,581) was a little below the previous year's total (1,784), which was the second-highest achieved in the last 10 years.

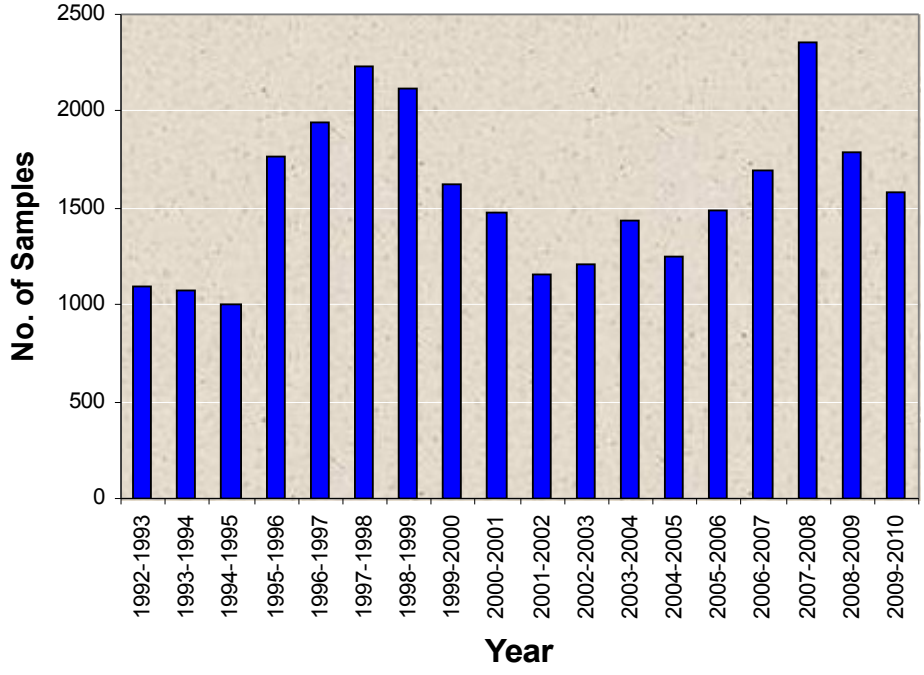
Background: Prior to 1992, the routine sample processing was carried out at Dwellingup Research Centre. Between 1984 and 1992 this was done by Janet Webster. All identification of *Phytophthora* species isolated from these samples between 1985 and 1992 was completed by Mike Stukely at Kensington. Records from these earlier years, for which map references are available, have been included in the **VHS database** which now contains a total of 34,951 records.

The VHS at Kensington was managed between 1992 and 2001 by Francis Tay, and from 2001 by Mike Stukely. Currently, Technical support (1 FTE) is provided on a time-share basis by Janet Webster and Juanita Ciampini, with additional casual assistance as required. The long-term continuous involvement by these staff members in this specialised laboratory role, and in assisting VHS clients, is a definite advantage.

Since 2005, a strong collaborative relationship has been developed with the **Centre for *Phytophthora* Science and Management** (CPSM) at Murdoch University (Director: Associate Professor Giles Hardy). This has enabled the testing by DNA sequencing of new as well as historical *Phytophthora* isolates, which has led to the discovery of the new, undescribed taxa and hybrids, and of new records for WA of several species and undescribed taxa that are known from elsewhere (**Section 3**). A student project investigating several of the new Phytophthoras is in progress at Murdoch University under joint supervision by Prof. Hardy, Dr Treena Burgess and M Stukely (**Section 6**).

Table 4 and Figure 4. Numbers of samples processed by the VHS for *Phytophthora* detection from 1992-1993 to 2009-2010.

Year	No. of Samples	Year	No. of Samples
1992-1993	1095	2002-2003	1210
1993-1994	1075	2003-2004	1435
1994-1995	1001	2004-2005	1250
1995-1996	1767	2005-2006	1486
1996-1997	1944	2006-2007	1696
1997-1998	2227	2007-2008	2353
1998-1999	2115	2008-2009	1784
1999-2000	1626	2009-2010	1581
2000-2001	1477		
2001-2002	1154	**TOTAL**	**28,276**



5. Concluding Comment

Land managers are encouraged to make full use of the services provided by the VHS for the detection of *Phytophthora*. The sample-processing service is provided free of charge to all DEC personnel and sections, and DEC-sponsored community groups. No other service that is capable of processing the required numbers of samples is available in WA.

The implementation of **Best Practice methods and standards** for managing *Phytophthora* dieback is based upon the key steps of detection, diagnosis, demarcation and mapping of infested areas, and hence the identification of uninfested areas and those that are **protectable** (CALM, 2002). The VHS laboratory's testing of samples for the presence of *Phytophthora* is an integral and essential part of this process.

It is important to recognise that areas of land must be **regularly re-assessed and re-tested** for *Phytophthora* infestation, since with time the pathogen will continue to spread from its known, established foci. This spread may be autonomous (by root-to-root contact between host plants, and through dispersal of zoospores in water), or through the activity of vectors such as native and feral animals, and people with their vehicles and machinery.

The **appropriate frequency of re-assessment and re-testing** for a given area of land will depend upon several factors:

- the **values** associated with that area,
- the **likelihood** or **level of risk** of introduction of **any** *Phytophthora* into that area, and
- the **consequences** of the introduction of **any** *Phytophthora* species to the ecosystem.

Information on the distribution of <u>all</u> *Phytophthora* species must be up-to-date, for land management to be most effective.

6. Collaborations and Student Projects (Co-supervised)

a. PhD project (Alex Rea) – **Classical and molecular taxonomy and pathogenicity testing of *Phytophthora* species** – commenced in April 2007 at Murdoch University.

b. CPSM project – Investigation of *Phytophthora* hybrids from WA natural ecosystems.

7. Recent Publications and Presentations by VHS Staff

- Research paper on the undescribed *Phytophthora,* 'P.sp.2', published. Stukely *et al*. (2007a): **A new homothallic *Phytophthora* from the jarrah forest in Western Australia**. *Australasian Plant Disease Notes* **2:** 49-51.
- Research paper on a new *Phytophthora* record for WA published. Stukely *et al*. (2007b): ***Phytophthora inundata* from native vegetation in Western Australia**. *Australasian Plant Pathology* **36:** 606-608.
- Oral Presentation to WA Dieback Information Group (DIG) meeting, 4 July 2008. Stukely: **New *Phytophthora* species in WA.**
- Poster presented at international conference. Stukely *et al*. (2007c): **Molecular testing uncovers new *Phytophthora* taxa from natural ecosystems in Western Australia.** 4th IUFRO Meeting on Phytophthoras in Forests and Natural Ecosystems, Monterey, California, USA, 26-31 August 2007.
- DEC Internal Report – a comprehensive review of the role of the VHS, its outputs, staff and facilities. Stukely (2008): **The Science Division's Vegetation Health Service – A Review, April 2008.**
- Paper prepared for international workshop. Stukely *et al*. (2008): **Molecular re-evaluation of *Phytophthora* taxa collected over the past three decades from natural ecosystems in Western Australia**. 3rd International *Phytophthora* and *Pythium* Workshop: "Integration of traditional and modern approaches for investigating the taxonomy and evolution of *Phytophthora*, *Pythium* and related genera" 23-24 August 2008, Turin, Italy (associated with the **International Congress of Plant Pathology**).

- Paper published in international journal. Burgess TI, Webster JL, Ciampini JA, White D, Hardy GEStJ, Stukely MJC (2009): **Re-evaluation of *Phytophthora* species isolated during 30 years of vegetation health surveys in Western Australia using molecular techniques.** *Plant Disease* **93**:215-223.
- Paper published in international journal. Scott PM, Burgess TI, Barber PA, Shearer BL, Stukely MJC, Hardy GEStJ, Jung T (2009): ***Phytophthora multivora* sp. nov., a new species recovered from declining *Eucalyptus*, *Banksia*, *Agonis* and other plant species in Western Australia. *Persoonia* 22:1-13.**
- DEC Science Division Information Sheet #8/2009. Stukely M (2009): **Phytophthora Dieback – detecting the pathogen**.
- Papers (2) presented at international conference. A paper on the new ***Phytophthora* taxa *P.*sp.2, *P.*sp.1 and *P.*sp.9**, and a second paper on **the occurrence in WA of unique *Phytophthora* hybrids**, were presented (by Dr T Burgess, CPSM) at the **5th IUFRO Conference, "*Phytophthora* in Forests and Natural Ecosystems"**, Rotorua, New Zealand, 7-12 March 2010.
- The formal descriptions of several of the new WA *Phytophthora* species are to be submitted for publication in 2010.

8. Other References Cited

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Maseko B, Burgess TI, Coutinho TA, Wingfield MJ (2007). Two new *Phytophthora* species from South African *Eucalyptus* plantations. *Mycological Research* **111**:1321-1338.

Stukely MJC, Shearer BL, Tay FCS, Hart RM, Hart RP (1997). *Phytophthora* species in natural vegetation in Western Australia. In: 'Programme and Summaries', 11th Biennial Conference, Australasian Plant Pathology Society, Perth, Western Australia, p.199. (Poster).

9. Attachments, Notes and Acknowledgments

1. **Map showing results of *Phytophthora* sample testing by the DEC Vegetation Health Service, 2009-2010.**

   Phytophthora cinnamomi (CIN), Other *Phytophthora* species (PSP), and Negative (NEG) recoveries are shown separately.

   The map was produced by Holly Smith (DEC Geographic Information Services).

2. **Phylogenetic trees (based on ITS rDNA sequences) showing a limited selection of the nearly 100 known species of *Phytophthora*, with the ten undescribed WA taxa, and five additional unique isolates in Clade 6 (indicative only, July 2008).**

   Tree 1: The ten undescribed taxa isolated from WA samples ("**P. spp. 1-11**") are highlighted red. The species groupings (Clades) proposed by Cooke *et al*. (2000) (Clades 1-9) are shown at the right.

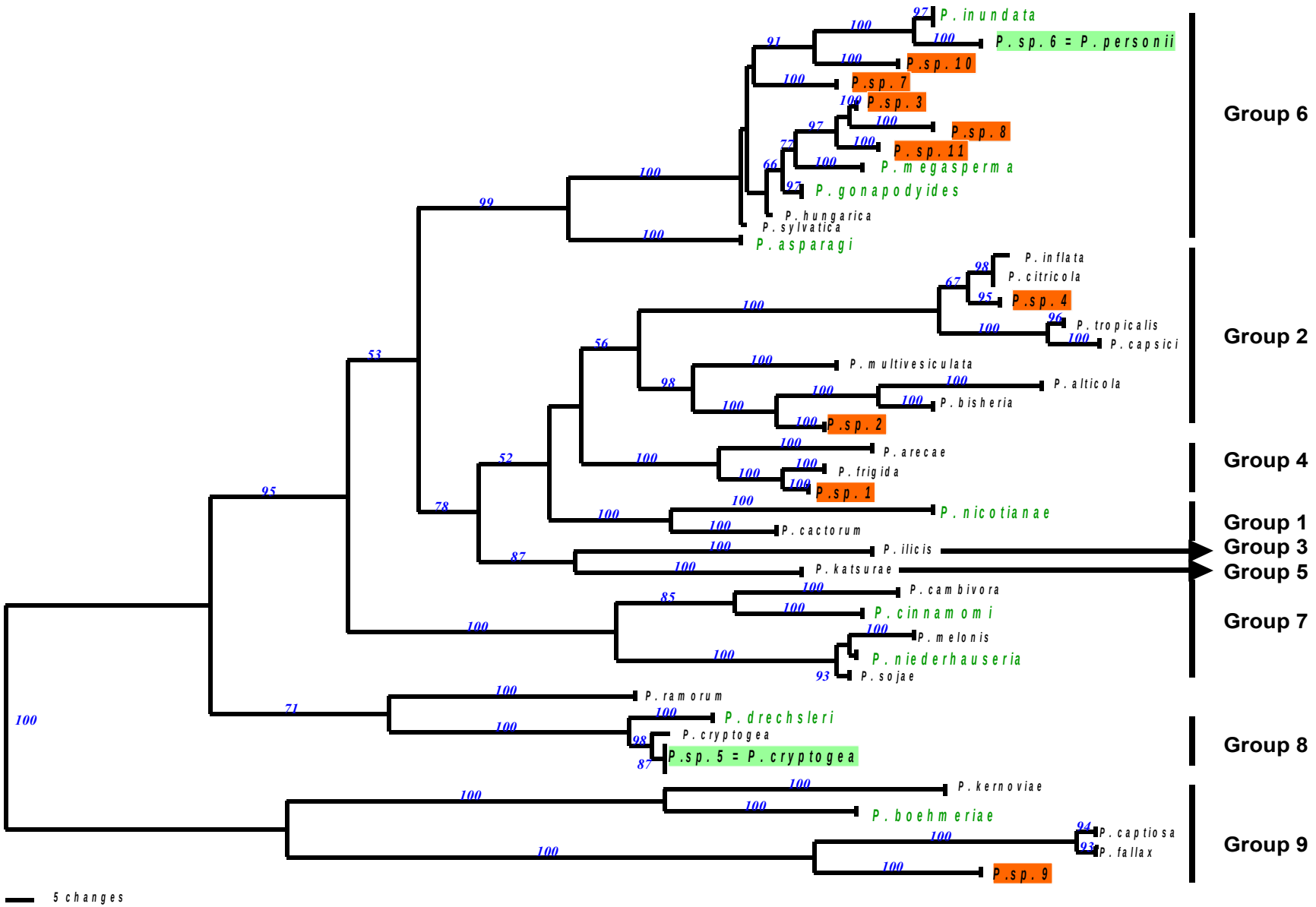
   Phytophthora species shown in green text are those believed previously to be present in WA natural ecosystems, based on the traditional morphological identification of pure cultures (Stukely *et al*. 1997; D'Souza *et al*. 1997).

   The other species shown include all of those most closely related to each of the 'new' WA taxa, based on analysis of their ITS rDNA sequences, from the international **GenBank database**.

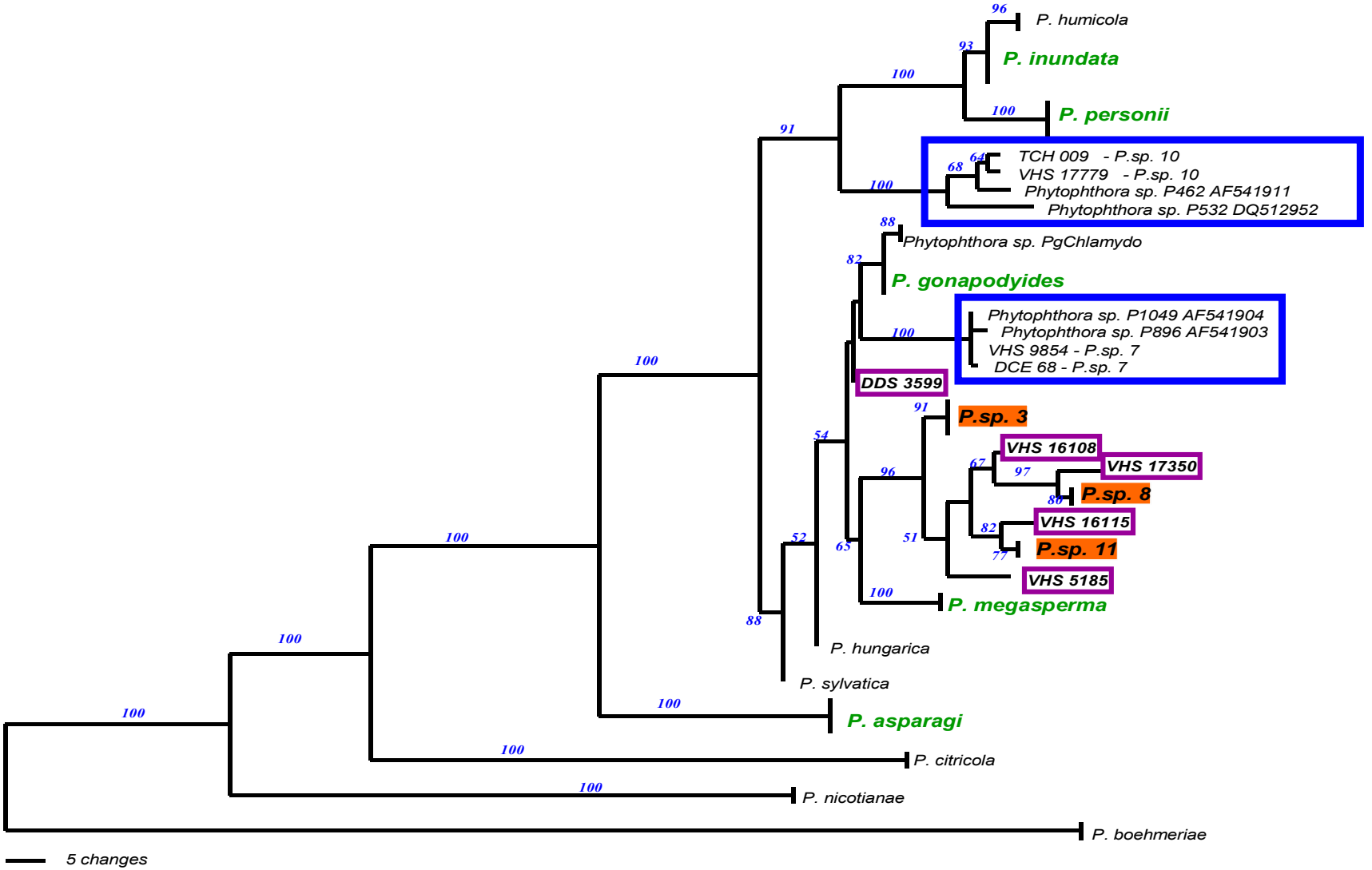
   Four newly-described *Phytophthora* species causing declines of eucalypts, in South Africa (*P. alticola, P. frigida* – Maseko *et al*. 2007) and New Zealand (*P. captiosa, P. fallax* – Dick *et al*. 2006), are also shown.

Tree 2: Detail of the new WA taxa in Cooke's Clade **6** (P.spp. 10, 7, 3, 8, 11), with their closest known relatives (see blue boxes). Five **unique** WA isolates are shown in purple boxes – new evidence (2009-2010) shows that four of these (and several others) are **hybrids**.

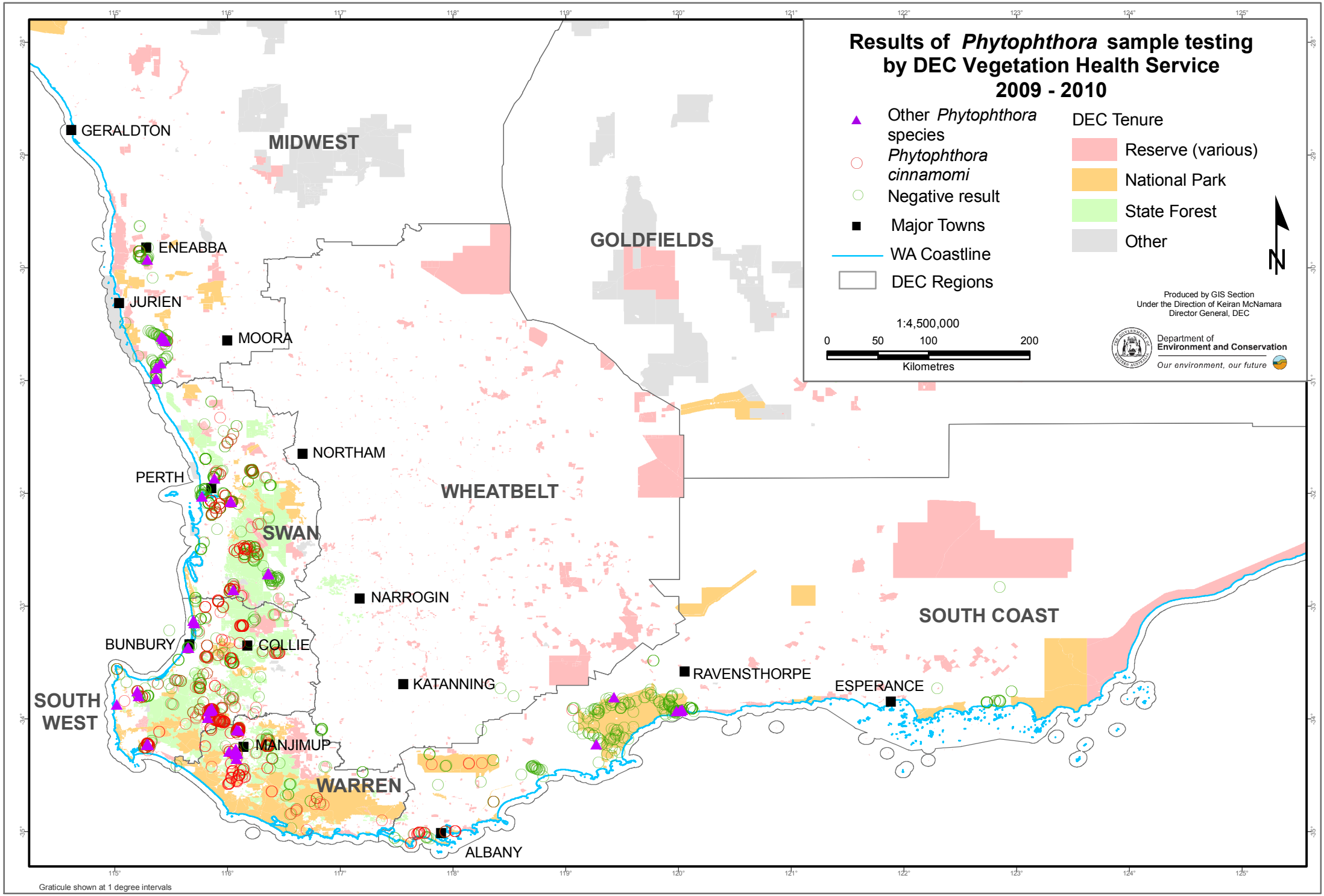
Analyses of rDNA sequence data and the construction of the phylogenetic trees were carried out by Dr Treena Burgess (CPSM, Murdoch University).



Phylogenetic Tree 1 – *Phytophthora* Clades (Groups) 1-9 (Cooke): New taxa 'P. spp. 1-11' are highlighted red. *VHS Annual Report 2009-2010.*



Phylogenetic Tree 2 – *Phytophthora* Clade (Group) 6 (Cooke): New taxa 'P. spp. 10, 7, 3, 8, 11'; and five unique isolates (see purple boxes).
VHS Annual Report 2009-2010.


Results of *Phytophthora* sample testing by DEC Vegetation Health Service 2009 - 2010
Other *Phytophthora* species
Phytophthora cinnamomi
Negative result
Major Towns
WA Coastline
DEC Regions
DEC Tenure
Reserve (various)
National Park
State Forest
Other
N
1:4,500,000
0 50 100 200
Kilometres
Produced by GIS Section
Under the Direction of Keiran McNamara
Director General, DEC
Department of Environment and Conservation
Our environment, our future
GERALDTON
ENEABBA
JURIEN
MOORA
NORTHAM
PERTH
NARROGIN
BUNBURY
COLLIE
KATANNING
RAVENSTHORPE
ESPERANCE
MANJIMUP
ALBANY
MIDWEST
GOLDFIELDS
WHEATBELT
SWAN
SOUTH COAST
SOUTH WEST
WARREN
Graticule shown at 1 degree intervals
Produced: 22/10/2010 Job Ref: G:\Projects\08122C04_Phytophthora_Mapping\2010\MXD