Department of Environment and Conservation

Vegetation Health Service

Annual Report 2010-2011

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 36,889 records (at 30th June 2011), 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 985 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 2010-2011 financial year, the VHS received 1,936 samples for testing for the presence of *Phytophthora* (**Table 1** and **Figure 1**). This was well above the previous year's total of 1,581 and is the second-highest yearly total processed in the last 12 years (see **Table 4** and **Figure 4**). The sources of the 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 (private consultants, land managers or owners).

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)**.

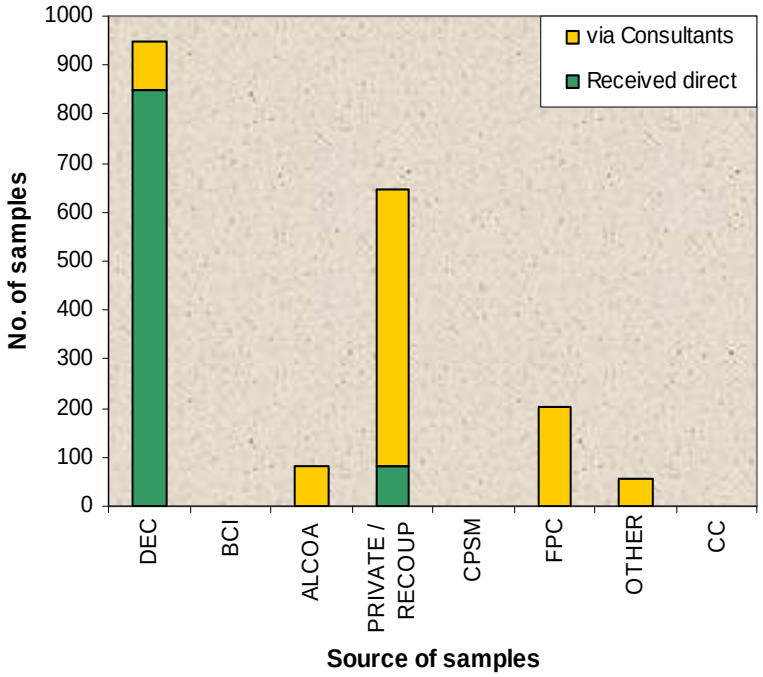
CPSM – samples associated with projects carried out at the **Centre for *Phytophthora* Science and Management** at Murdoch University.

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**). A slightly smaller number of Consultants – Glevan Consulting, Dieback Treatment Services, NPC Consulting, Moore Mapping, NRG Consulting, RPS Environmental, Natural Area Consulting, Great Southern Biologic, and Ekologica – supplied 1,008 samples from various sources including from DEC land and Alcoa (**Table 1** and **Figure 1**). Alcoa World Alumina Australia was the only mining company sending samples direct to the VHS this year. No samples were received from Caring for our Country (formerly NHT) projects in 2010-11. 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, Colin Crane).

Table 1 and Figure 1. Numbers of samples received by the VHS from major sources, including consultants, in 2010-2011. Numbers of samples received from the same sources in 2009-2010 are shown in the Table for comparison. Abbreviations of sources are as listed above.

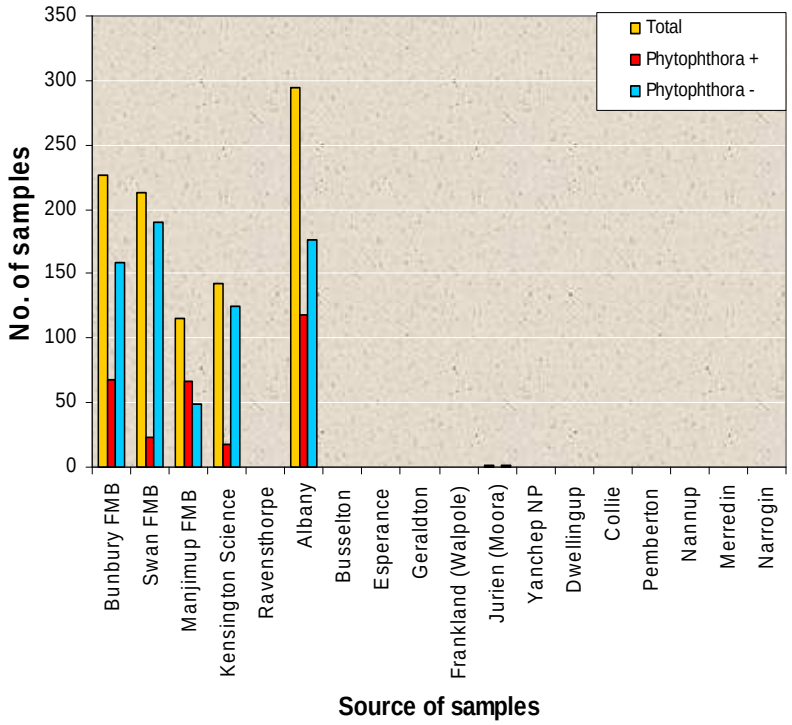
Source	No. of samples			
	Received direct	Received via Consultants	Total 2010-2011	Total 2009-2010
DEC	847	100	947	618
BCI	0	0	0	128
ALCOA	0	84	84	112
PRIVATE / RECOUP	81	565	646	529
CPSM	0	0	0	0
FPC	0	204	204	131
OTHER	0	55	55	63
CC	0	0	0	0
TOTAL	**928**	**1008**	**1936**	**1581**



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 into three categories to distinguish *P. cinnamomi*, Other *Phytophthora* spp. (including undescribed new *Phytophthora* taxa), and Negative.

Table 2 and Figure 2. Numbers of samples received by the VHS from DEC offices in 2010-2011, and numbers giving positive or negative recoveries of *Phytophthora*. Total numbers of samples received from the same sources in 2009-2010 are shown in the Table for comparison. DEC samples received from consultants are not included.

DEC Office	No. of Samples			
	Total received 2010-2011	*Ph*. Positive 2010-2011	*Ph*. Negative 2010-2011	Total received 2009-2010
Bunbury FMB	227	68	159	384
Swan FMB	213	23	190	32
Manjimup FMB	115	66	49	146
Kensington Science	142	17	125	19
Ravensthorpe	0	0	0	0
Albany	294	118	176	81
Busselton	0	0	0	0
Esperance	0	0	0	2
Geraldton	0	0	0	0
Frankland (Walpole)	0	0	0	20
Jurien (Moora)	2	0	2	0
Yanchep NP	0	0	0	0
Dwellingup	0	0	0	0
Collie	0	0	0	0
Pemberton	0	0	0	0
Nannup	0	0	0	0
Merredin	0	0	0	0
Narrogin	0	0	0	0
TOTAL	**991**	**292**	**699**	**684**



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 morpho-species (in addition to *P. cinnamomi*) had previously been documented in association with dying plants 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 international authorities such as CABI (UK) and CBS (The Netherlands) for verification.

In addition to the above species, ***P. inundata*** had been identified in WA by DNA sequencing prior to June 2007 (Stukely *et al*., 2007b). Testing of new and historical isolates by this technique since 2005 has led to the discovery of as many as twelve **new and previously undescribed *Phytophthora* species** in WA natural ecosystems. These were initially designated "**P.sp.1–12"**, and were mostly associated with dying vegetation (see **Section 3.1**) (Stukely *et al*., 2007c; Stukely *et al*., 2008; Burgess *et al*., 2009). In addition to ***Phytophthora multivora***, a further eight of these new species have now been described: ***P. elongata, P. thermophila, P. gibbosa, P. gregata, P. litoralis, P. arenaria, P. constricta*** **and** ***P. fluvialis.*** References are given in **Section 7** (p. 12). Pathogenicity has so far been tested and confirmed on native plants for *P. multivora*, *P. elongata, P. arenaria* and *P. constricta*. Several additional new WA taxa await formal description. New records for WA of several *Phytophthora* taxa that are known from elsewhere have also been discovered, and these are listed in **Section 3.1** (p.7).

During 2010-2011, *Phytophthora* was isolated from 590 of a total of 1,936 samples processed (compared with 507 of 1,581 in 2009-2010) (**Table 3, Figure 3**). ***Phytophthora cinnamomi*** was, as usual, the species most frequently isolated (544 samples, compared with 443 in 2009-2010). ***P. cryptogea*** was isolated from 19 samples, ***P. multivora*** from 12, ***P. inundata*** from 4, and ***P. nicotianae*** from 4. Two samples gave isolates that were shown by DNA sequencing to be members of other new (as yet undescribed) *Phytophthora* taxa (see **Section 3.1**): P. taxon rosacearum-like (1), and a unique hybrid (1). Another five *Phytophthora* isolates, from some of the most recent samples tested, were still to be fully identified to species.

One sample yielded both *P. cinnamomi* and *P. cryptogea*.

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 undescribed *Phytophthora* taxa); and c). Negative.

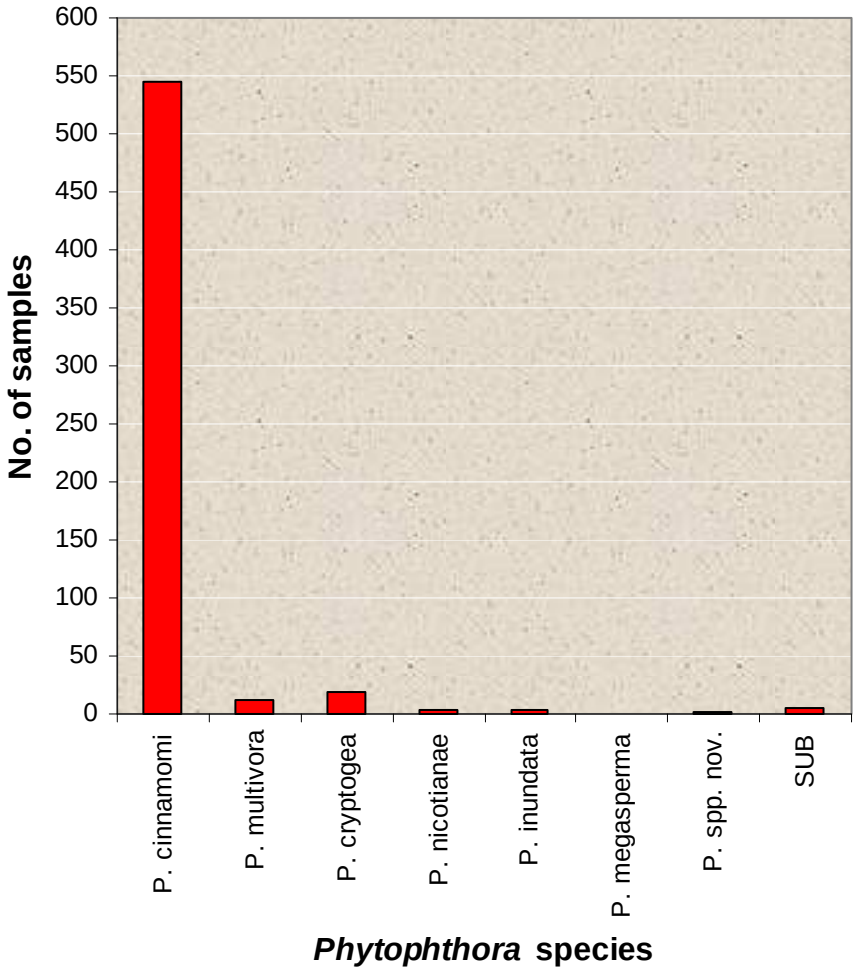
Representative pure living cultures of all *Phytophthora* species isolated, and of individual species representing different geographic locations, ecosystems, or host plants, or morphological types, were added to the permanent **VHS Culture Collection**. These cultures are maintained in a pure and viable condition by periodically sub-culturing, checking their purity and establishing fresh storage cultures. From this year, Hemp seed is now progressively being introduced as the main storage medium. There are now 985 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). Various backup cultures are also held. 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 2010-2011. Corresponding numbers of samples for 2009-2010 are shown in the Table for comparison.

***Phytophthora* species**	**No. of samples with positive *Phytophthora* recovery 2010-2011**		**No. of samples with positive *Phytophthora* recovery 2009-2010**	
	Primary result	**Second species[1]**	**Primary result**	**Second species[1]**
P. cinnamomi	544		443	
P. multivora[2]	12		38	4
P. cryptogea	19	1	7	
P. nicotianae	4		1	
P. inundata	4		2	
P. megasperma	0		0	
P. spp. nov.[3]	2		16	
SUB[4]	5		0	
TOTAL	**590**		**507**	**4**

[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, including hybrids (see Section 3.1).
[4]SUB – these *Phytophthora* cultures are still in the process of being identified.



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, with well over 50 new species having been discovered, worldwide, since 2000. To put this into perspective, a total of just 55 species had been formally described up to 1999. 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 564 recent and historical isolates of *Phytophthora* (to 30 June 2011), 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 database of DNA sequences that was created in 1988 and is in the public domain.]

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

- There are now known to be as many as **twelve previously undescribed new species of *Phytophthora* present in natural ecosystems in WA**.
- 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. 13).
- Nine of the new WA species have now been formally described: ***P. multivora, P. elongata, P. thermophila, P. gibbosa, P. gregata, P. litoralis, P. arenaria, P. constricta*** and ***P. fluvialis.*** Several additional new WA taxa await formal description.
- Pathogenicity has been tested and confirmed on selected WA native plant hosts for ***P. multivora, P. elongata, P. arenaria*** **and** ***P. constricta*** (Murdoch University projects).
- Two of these species, *P. arenaria* and *P. constricta*, are believed to be endemic in WA.
- New records for WA, of *Phytophthora* taxa that are known elsewhere, have included: ***P. inundata, P. niederhauserii, P.*** **taxon asparagi, *P.*** **taxon personii, *P.*** **taxon PgChlamydo, *P.*** **taxon rosacearum-like, *P.*** **taxon salixsoil** and ***P.*** **taxon humicola-like.**
- In addition to the above new species and records (for each of which there are multiple isolates), there are several unique ***Phytophthora*** **hybrid** isolates from natural ecosystems in WA **(Burgess *et al.* 2010)**. Larger numbers of hybrids have been isolated from WA waterways in the "Fishing for *Phytophthora*" project at CPSM. Further investigation of these *Phytophthora* hybrids and their origins is in progress. The number and diversity of the *Phytophthora* hybrids found in WA represents a unique situation worldwide, to date.
- The previously recorded presence in WA natural ecosystems of some *Phytophthora* species other than *P. cinnamomi* (***P. cryptogea, P. nicotianae, P. megasperma, P. boehmeriae***) has been confirmed by DNA sequencing of stored isolates; however, ***P. citricola*** and ***P. drechsleri*** (previously believed to be present, based on morphology) are in fact not present among the isolates tested to date.
- Still only **two WA isolates** of the many identified earlier as ***P. megasperma*** (based on their morphology) have had a 100% match to DNA sequences for *P. megasperma* on GenBank. The other isolates represent several diverse new taxa.
- Many (but not all) of the nine newly-described *Phytophthora* species, and all of the unique isolates, have so far been recorded **only in WA**.

- All but one (*P. fluvialis*, 'Psp8') of the nine newly-described *Phytophthora* species have been found **associated with dying indicator plants** in WA native forest or heath-land.
- Several of the new *Phytophthora* taxa are associated with deaths of native WA plants representing multiple families.
- The number of plant species associated with the new Phytophthoras is still increasing rapidly, as new isolates are found.
- Some *Phytophthora* taxa appear to have limited distribution, while others like *P. multivora* are widespread with an overall range similar to that of *P. cinnamomi*. However, some Phytophthoras may be active in a broader range of site conditions than those favouring *P. cinnamomi* (e.g. *P. multivora* in limestone soils, affecting tuart).

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

Many of the previously 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 previously undescribed Phytophthoras are all properly described and documented as separate, individual components of our biodiversity.

The previously undescribed Phytophthoras, and those newly recorded in WA, 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 completely "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. Pathogenicity has been tested and confirmed on selected native plant hosts for *P. multivora*, *P. elongata*, *P. arenaria* and *P. constricta*.

- Some (but not all) of the new WA *Phytophthora* taxa are likely to be **indigenous**.
- Several unique *Phytophthora* **hybrids** are also present.

Some of the new Phytophthoras (eg *P. multivora*, formerly identified as '*P. citricola'*), <u>but not all</u>, are widespread and are associated mostly with low-impact dieback sites. **However, it is important at this stage that they should <u>not</u> all be treated as indigenous (and hence assumed to be "not threatening"), until they have been investigated further.** It appears that they are not particularly new introductions, with some isolates of most species 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 has been known since the 1980s that, for example, 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 new 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.

All of the new *Phytophthora* taxa discovered in WA so far are **soil-borne**, so the current, proven approaches and procedures being used to manage *P. cinnamomi* are appropriate at this stage for managing all of them.

Research projects (**Section 6**) are providing 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.

However, it is important to note that the controls applied currently for *Phytophthora cinnamomi* (such as phosphite application, forest hygiene, and restricting operations to "dry soil" conditions) may not always be appropriate, nor directly applicable without modification, for managing some potential 'new' *Phytophthora* species, eg those now causing severe tree disease overseas, but not yet present here (*P. ramorum*, *P. kernoviae*). These two species produce **air-borne** spores and represent major biosecurity risks for Australia.

In relation to biosecurity, it should also be noted that four other newly-described *Phytophthora* species have recently been reported as causing disease in plantation eucalypts overseas: *P. captiosa* and *P. fallax* in New Zealand (Dick *et al*. 2006) and *P. alticola* and *P. frigida* in South Africa (Maseko *et al*. 2007). Of these four species only *P. fallax* has been found in Australia (Victoria), but not in association with diseased plants.

The newly discovered WA ***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, through the contact and interaction of compatible Phytophthoras, 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 30,212 samples have been processed for *Phytophthora* detection (to 30th June 2011) (**Table 4** and **Figure 4**). The 2010-2011 total (1,936) was well above the previous year's total (1,581), and is the second-highest achieved in the last 12 years.

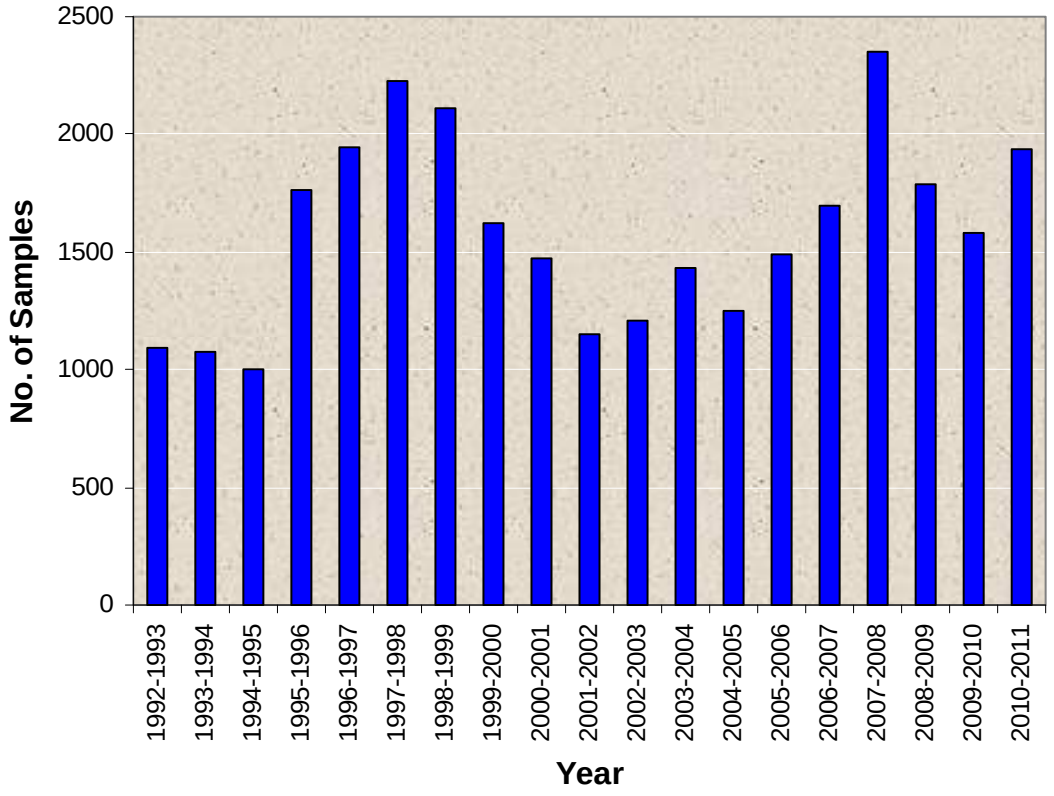
Background: Prior to 1992, the routine sample processing was carried out at Dwellingup Research Centre. Between 1984 and 1992 this was done by Janet Webster. All identification of *Phytophthora* species isolated from these samples between 1985 and 1992 was completed by Mike Stukely at Kensington. Records from these earlier years, for which map references are available, have been included in the **VHS database** which now contains a total of 36,889 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, previously undescribed species, the new records for WA of several species and undescribed taxa that are known from elsewhere, and the hybrids (**Section 3**). A student project investigating several of the new Phytophthoras is nearing completion 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 2010-2011.

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	2010-2011	1936
2001-2002	1154	**TOTAL**	**30,212**



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 the re-assessment and re-testing** for a given area of land will depend upon several factors, including:

- 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. [An update will be presented at DIG 2011.]**
- 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**. DEC Science Division.
- 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.
- Paper published in international journal. Rea AJ, Jung T, Burgess TI, Stukely MJC, Hardy GE St J (2010) ***Phytophthora elongata*** **sp. nov. a novel pathogen from the *Eucalyptus marginata* forest of Western Australia.** *Australasian Plant Pathology* **39,** 477-491.
- Paper published in international journal – descriptions of *P. thermophila, P. gibbosa, P. gregata, P. litoralis.* Jung T, Stukely MJC, Hardy GE St J, White D, Paap T, Dunstan WA, Burgess TI (2011) **Multiple new *Phytophthora* species from ITS clade 6 associated with natural ecosystems in Australia: evolutionary and ecological implications.** *Persoonia* **26,** 13-39.
- Paper published in international journal – descriptions of *P. arenaria* and *P. constricta*. Rea AJ, Burgess TI, Hardy GE St J, Stukely MJC, Jung T (2011) **Two novel and potentially endemic species of *Phytophthora* associated with episodic dieback of Kwongan vegetation in the south-west of Western Australia.** *Plant Pathology* Doi: 10.1111/j.1365-3059.2011.02463.x
- Paper published in international online journal – Jung T, Burgess TI, Hüberli D, Hardy GEStJ, Stukely M (2011) ***Phytophthora fluvialis*** **T. Jung & T.I. Burgess, *sp. nov*.** *Persoonia* **26**:146-147.

8. Other References Cited

CALM (2002) ***Phytophthora cinnamomi*** **and disease caused by it. Volume I – Management Guidelines.**

Cooke DEL, Drenth A, Duncan JM, Wagels G, Brasier CM (2000) **A molecular phylogeny of *Phytophthora* and related oomycetes.** *Fungal Genetics and Biology* **30**: 17-32.

Dick MA, Dobbie K, Cooke DEL, Brasier CM (2006) ***Phytophthora captiosa*** **sp. nov. and *P. fallax* sp. nov. causing crown dieback of *Eucalyptus* in New Zealand.** *Mycological Research* **110**: 393-404.

D'Souza NK, Webster JL, Tay FCS (1997) ***Phytophthora boehmeriae*** **isolated for the first time in Western Australia.** *Australasian Plant Pathology* **26**: 204.

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, 2010-2011.**

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

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

2. **Phylogenetic trees (based on ITS rDNA sequences) showing the newly described WA *Phytophthora* species and other new records, and additional unique hybrid isolates in Clade 6 (indicative only, 2011).**

   Tree 1: A limited selection of the formally described species of *Phytophthora* worldwide (the total number of known species is now well over 100), showing the species groupings (**Clades 1-10**) proposed by Cooke *et al*. (2000) at the right. Four newly-described WA species (***P. arenaria, P. elongata, P. multivora, P. constricta***) are shown in larger black print, with their earlier "Psp" numbers. Those shown in blue text are **new records for WA** of taxa known from elsewhere. *Phytophthora* species shown in red text are those believed previously to be present in WA natural ecosystems, based on the traditional morphological identification of pure cultures.

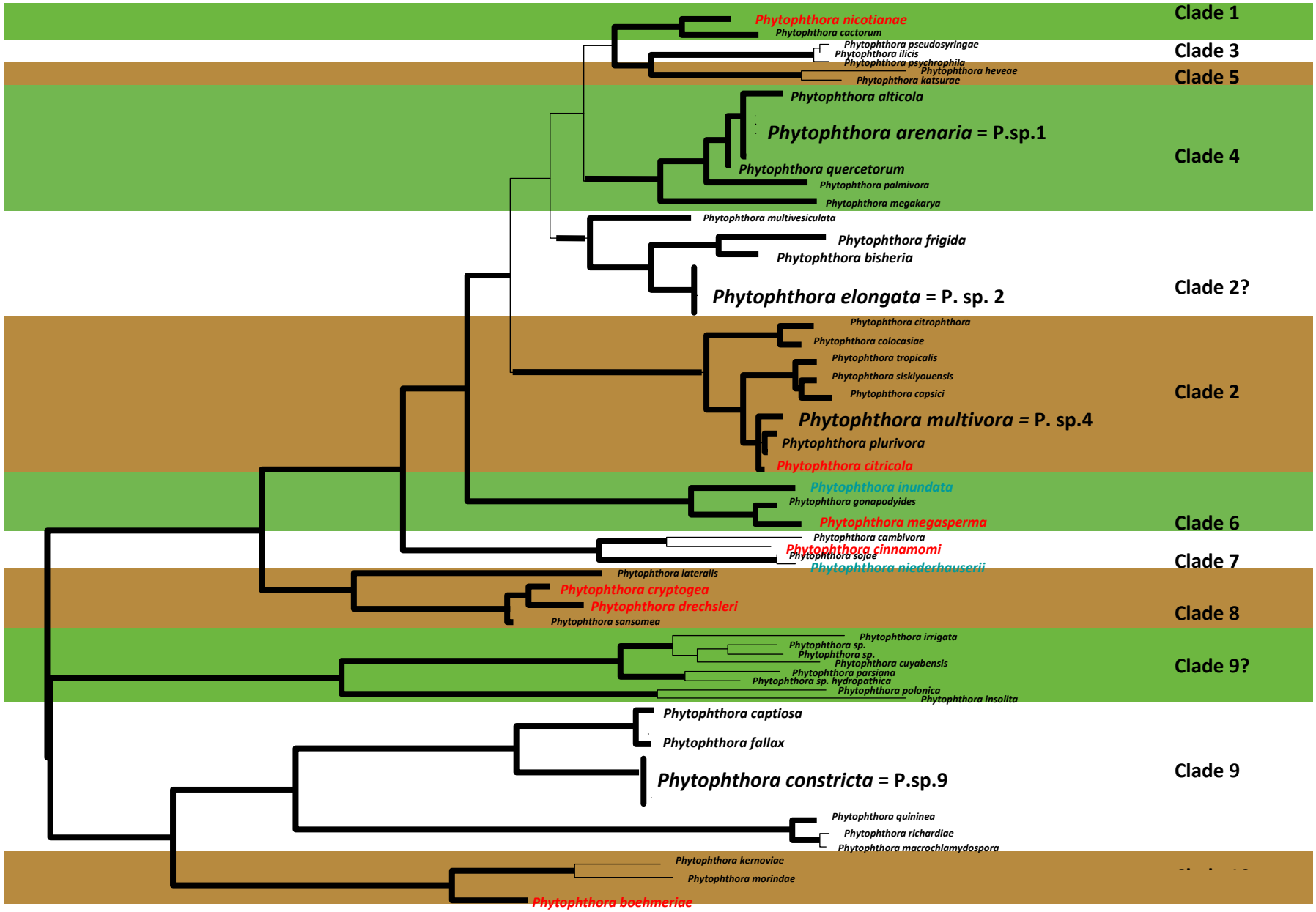
   The other species shown include all of those most closely related to each of the new WA taxa, based on analysis of their ITS rDNA sequences, from the international **GenBank database**.

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

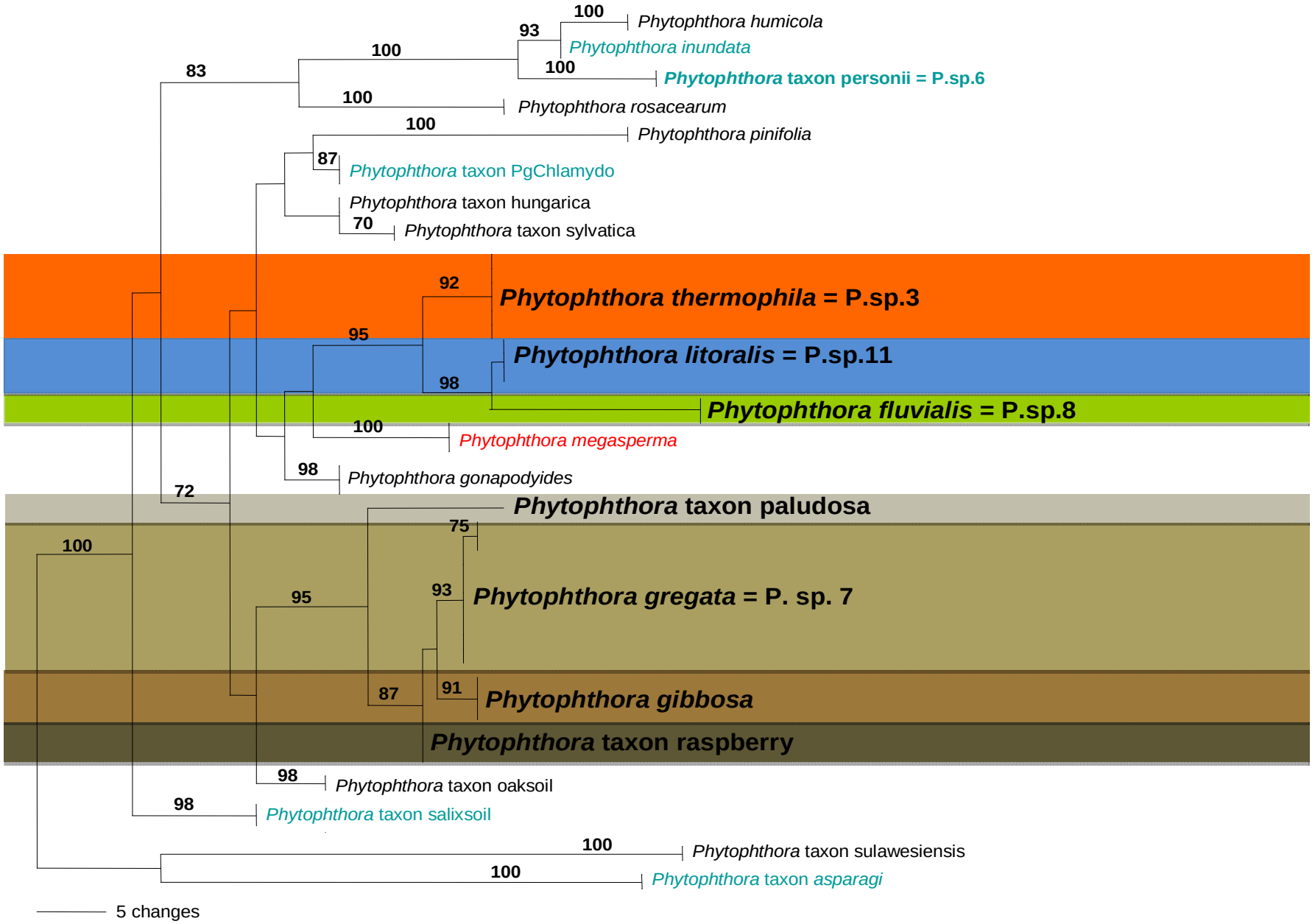
   Tree 2: Cooke's **Clade 6,** showing five newly-described WA *Phytophthora* species (***P. thermophila, P. litoralis, P. fluvialis, P. gregata, P. gibbosa***) in larger black print, with their earlier "Psp" numbers, and with their closest known relatives. Those shown in blue text are **new records for WA** of taxa known from elsewhere. *P. megasperma* is shown in red text.

   Tree 3: Detail within Cooke's **Clade 6,** around the new WA species ***P. thermophila, P. litoralis*** and ***P. fluvialis.*** The numbers represent individual isolates, many of them **hybrids**. [VHS, DDS, HSA = VHS Collection; DH = "Fishing for *Phytophthora*" project, isolates from water (D Huberli, CPSM).]

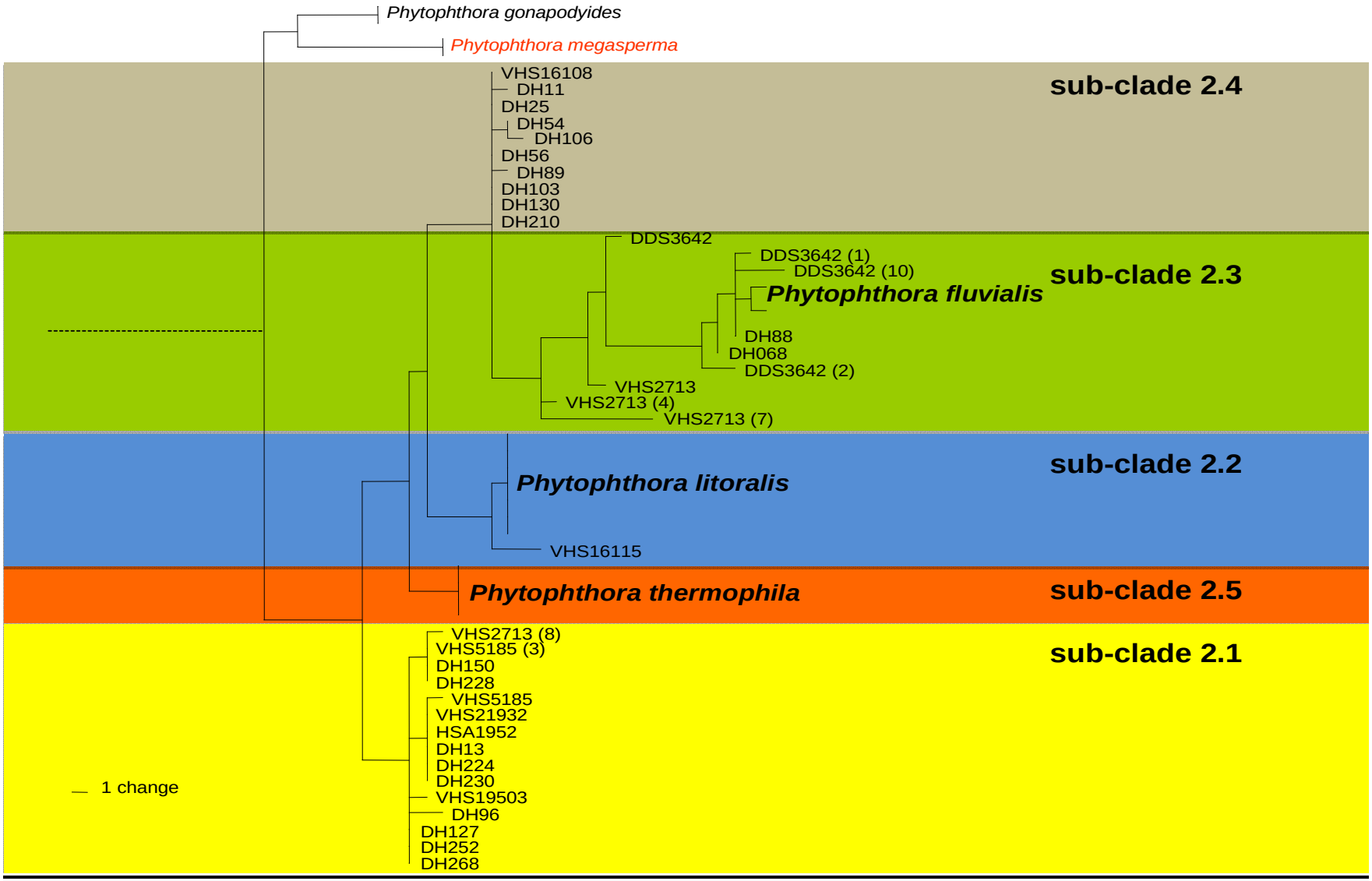
   Acknowledgment: Analyses of rDNA sequence data and the construction of the phylogenetic trees were carried out by Dr Treena Burgess and Diane White (CPSM, Murdoch University).



<u>Phylogenetic Tree 1</u> – Genus *Phytophthora*, showing selected species from Clades 1-10 (after Cooke *et al.* 2000); new WA species (large black font); new WA records (blue); and species previously believed present in WA (red). *VHS Annual Report 2010-2011 (T Burgess)*

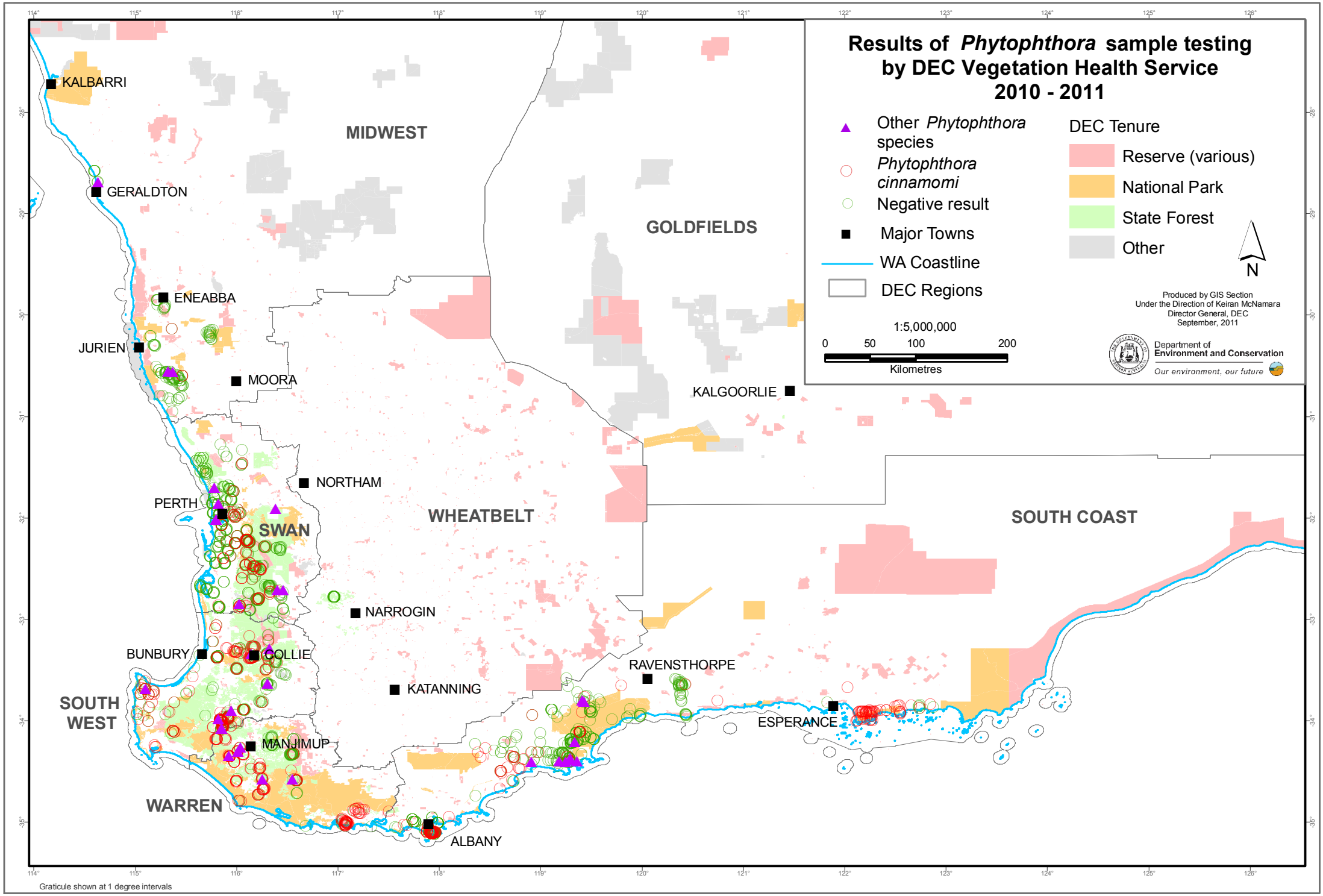


<u>Phylogenetic Tree 2</u> – Clade 6 (after Cooke *et al.* 2000) of genus *Phytophthora*, showing new WA and other species (large black font); new WA records (blue); and species previously believed present in WA (red). *VHS Annual Report 2010-2011 (T Burgess)*



<u>Phylogenetic Tree 3</u> – Detail within Clade 6 (after Cooke *et al.* 2000) of genus *Phytophthora*, showing three new WA species (large black font); and individual hybrid isolates (VHS, DDS, HSA isolates are from the VHS collection; DH are from the "Fishing for *Phytophthora*" project at CPSM).

VHS Annual Report 2010-2011 (T Burgess)



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