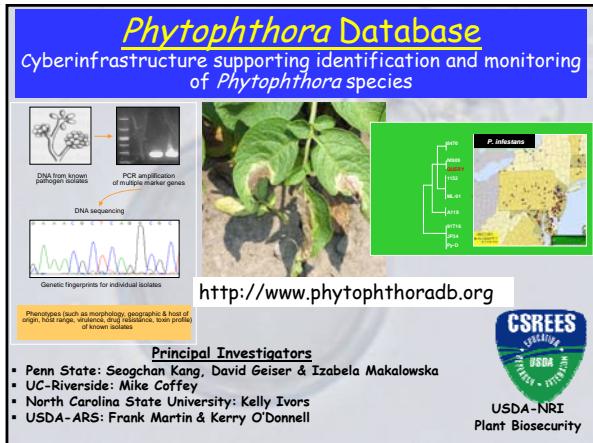


Phytophthora Database

Cyberinfrastructure supporting identification and monitoring of *Phytophthora* species

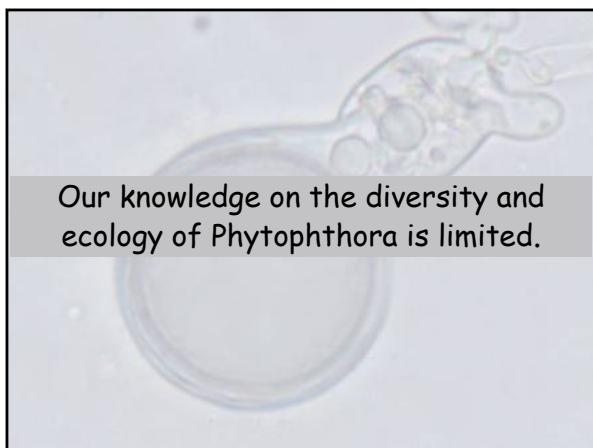


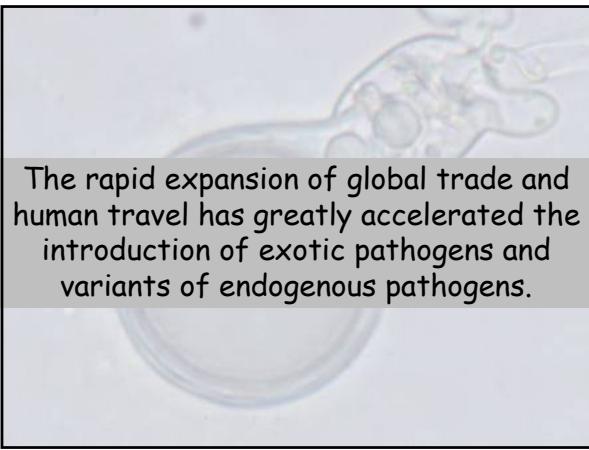
Data from known Phytophthora isolates
PCR amplification of unique marker genes
DNA sequencing
Genetic fingerprints for individual isolates
Phenotypes (such as morphology, geographic & host origin, host range, pathogenicity, virulence, toxic profile of known isolates)

<http://www.phytophthoradb.org>

Principal Investigators
• Penn State: Seogchan Kang, David Geiser & Izabela Makalowska
• UC-Riverside: Mike Coffey
• North Carolina State University: Kelly Ivors
• USDA-ARS: Frank Martin & Kerry O'Donnell

CSREES
USDA-NRI
Plant Biosecurity





The rapid expansion of global trade and human travel has greatly accelerated the introduction of exotic pathogens and variants of endogenous pathogens.

Objectives of the Project

- ✓ To establish a comprehensive phylogenetic framework for the genus *Phytophthora*
- ✓ To build an internet database that crosslinks the genotype, phenotype, and distribution of individual *Phytophthora* species/isolates (generate data for database)
- ✓ To develop and optimize molecular diagnostic tools for detecting and identifying *Phytophthora*

Park, J., Ivors, K., Kang, S. et al. 2008. *Phytophthora Database: A forensic database supporting the identification and monitoring of Phytophthora*. Plant Dis. 92:966-972.

Organization of the *Pytophthora* Database

www.PhytophthoraDB.org

The screenshot shows the homepage of the Phytophthora Database. At the top, there's a blue header bar with the title "Organization of the Pytophthora Database". Below it is a navigation menu with links like "Home", "Introduction", "Database", "Search & Analysis", and "Refs. & News". The main content area features a large image of a plant with visible root structures. On the left, there's a "Phytophthora Database Login" section with a "Dear User" message and a "Run Wizard" button. The right side has sections for "Welcome to Phytophthora Database", "Current Statistics of the Database" (listing 94 species, 3,772 isolates, and 3,080 references), and "Contact the Database Curator". Logos for USDA, APS PRESS, and KACC are at the bottom, along with a citation: "Park et al. Plant Disease (2008) 92: 966-972".

Species Information Page

Phytophthora DATABASE

This page provides detailed information about a specific species of Phytophthora. It includes a navigation menu, a search bar, and several sections of content.

Navigation:

- Home
- Introduction
- Database
- Search & Analyse
- Geographical Distribution

Species: *P. ramorum*

Identification: Click 1, Click 2, Click 3, Click 4

Image: C (Close-up image of tree trunk showing symptoms)

Text: Detailed description of the species, its biology, and its impact on various host plants.

Links: References, Further reading, and Related links.

Bottom: A link to "A full list of species in the genus Phytophthora".

Phytophthora DATABASE

www.Phytophtoradb.org

Species List

There are 83 species.

Species Name	Isolate Count	Function
<i>Peronophthora alni</i>	1	[Modify]
<i>Phytophthora aesculi</i>	3	[Modify]
<i>Phytophthora aculeata</i>	2	[Modify]
<i>Phytophthora aphanidis</i>	2	[Modify]
<i>Phytophthora aphyllae</i>	2	[Modify]
<i>Phytophthora aphyllae</i>	1	[Modify]
<i>Phytophthora aphyllae</i>	1	[Modify]
<i>Phytophthora brasiliensis</i>	5	[Modify]
<i>Phytophthora cinnamomi</i>	833	[Modify]
<i>Phytophthora corynorhiza</i>	1	[Modify]
<i>Phytophthora cambivora</i>	29	[Modify]
<i>Phytophthora capsici</i>	20	[Modify]
<i>Phytophthora capsici</i>	3	[Modify]
<i>Phytophthora cinnamomi</i>	75	[Modify]
<i>Phytophthora citrulls</i>	125	[Modify]
<i>Phytophthora clematiphila</i>	51	[Modify]
<i>Phytophthora clematiphila</i>	1	[Modify]
<i>Phytophthora clematiphila</i>	2	[Modify]
<i>Phytophthora clematiphila</i>	4	[Modify]
<i>Phytophthora corynorhiza</i>	0	[Modify]
<i>Phytophthora corynorhiza</i>	0	[Modify]
<i>Phytophthora dampicola</i>	120	[Modify]
<i>Phytophthora dampicola</i>	2	[Modify]
<i>Phytophthora dampicola</i>	3	[Modify]
<i>Phytophthora dampicola</i>	1	[Modify]
<i>Phytophthora dampicola</i>	4	[Modify]
<i>Phytophthora dampicola</i>	3	[Modify]
<i>Phytophthora dampicola</i>	4	[Modify]
<i>Phytophthora dampicola</i>	2	[Modify]
<i>Phytophthora dampicola</i>	3	[Modify]
<i>Phytophthora dampicola</i>	2	[Modify]

Page: 1 / 1

www.Phytophtoradb.org

Symptoms:

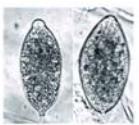
Because there are over 800+ host species for *P. ramorum* at current, there are many different types of disease symptoms associated with the pathogen.

Symptoms on true oak (*Quercus spp.*) and tanoak (*Lithocarpus densiflorus*) include blackened trunk / bark cankers, often with a tan or reddish discoloration. These cankers can girdle the trunk, causing the tree to die. On some oak species, trunk cankers are restricted to above the soil line. On many oak species, tree mortality often occurs, but usually not as sudden as the disease name suggests, because they may continue to live for up to one year after tree death.

Image: Tan oak canker (Close-up image of tree trunk showing symptoms)

Image: Tan oak canker (Close-up image of tree trunk showing symptoms)

Life Cycle and Morphology



Perithecioid conidial angiospermous germination of *P. ramorum*.

Known Diseases

Due to the high level of quarantine status of this pathogen, several detection techniques exist and many (indirect) protocols have been validated by the USDA-APHIS for use by state agencies. For a summary of procedures approved by APHIS, please refer to the LRU (http://www.aphis.usda.gov/lru).

Disease specific protocols for *P. ramorum* can be divided into three different types of methods:

1) Isolation of *P. ramorum* onto selective media, followed by a morphological identification of the pathogen microscopically. Most of the selective media used for *P. ramorum* isolation are amended with antibiotics, including thiomersal, Ampicillin, and DOWS.

2) Detection of *P. ramorum* via polymerase chain reaction (PCR), either real-time PCR or conventional PCR.

3) Substrate and Taqman real-time PCR in the ITS region (Hoyle et al., 26; Hughes et al., 36), and Taqman real-time PCR in the

Diseases

Phytophthora root rot, Phytophthora root rot caused by several *Phytophthora* spp., including *P. cinnamomi*, has been associated with significant damage to Fraser fir (Abies fraseri) in the southern Appalachian region since the 1980s. *Phytophthora* species represent a group of soil-borne fungi that are found throughout the world. They are found in all major forest types, including Douglas fir, white pine, and other species). However, annual losses due to *Phytophthora* are estimated at 1 million and long-term yield reductions of 10% are projected for the southern Appalachians over the next 50 years. *P. cinnamomi* has also been isolated from *Abies balsamea* roots and surrounding soils, although it does not account for most of the disease incidence in Fraser fir (Bennet & Thompson, 1992). *P. cinnamomi* is also known to cause root rot in eastern hemlock (Tsuga canadensis) and white spruce (Picea glauca) in the Adirondack mountains (Bennet & Grand, 2000). *P. cinnamomi* has been implicated in the decline of eastern hemlock in the Adirondacks, where the disease incidence is approximately 10% and mortality is 50% (Bennet & Grand, 2000). The incidence of *P. cinnamomi* in eastern hemlock in the Adirondacks is estimated to be 10% and mortality is 50% (Bennet & Grand, 2000). The percentage assesses small 10-m² transects to approximately 4 infected trees per hectare in a standard 1.8 to 3.5 hectare. Tree assessments at field transects may vary because of *Phytophthora* during periods that are favorable for disease development, resulting in further impacts to the growth and survival of healthy understory trees. After disease develops in the field, the disease may spread to other trees in the same stand, and may spread to other stands through seed dispersal. *P. cinnamomi* can cause chain diseases in the soil, in pieces of organic matter, or in roots of seedlings and trees. Due to this persistence, once the pathogen is established in a stand, it is very difficult to control. *P. cinnamomi* is a soil-borne pathogen that can survive in the soil for decades. Disease, such as yellow-green needles and dead branches, are not obvious until the roots are heavily colonized, after which death of the tree occurs within 1-2 years.



Life Cycle and Morphology

P. cinnamomi is capable of reproducing both sexually, which produces meiospores, and asexually, which produces sporangia. Sporangia are produced on the surface of the host plant and give macroscopically in size and can persist in soil or infected plant material for months, to some extent seasons.

When conditions favor asexual growth, the pathogen enters the asexual sporulation cycle. Myceliae will differentiate and produce sporangia, which then rupture and release 20–30 biflagellate zoospores (2 cells). These zoospores swim chemotactically toward host tissue and penetrate it. Once inside the host, the pathogen begins to grow and produce more mycelium. As the pathogen grows, more sporangia can be produced, thus repeating the asexual cycle and rapidly increasing levels of infection.



P. cinnamomi pear shaped (oblongiform) sporangium that is nonpolar and nonredundant.



P. cinnamomi mycelium showing dense growth (the clusters cover fungal fruits).

P. cinnamomi is heterothallic. Crossing from when A1 and A2 mating types are paired, although it has been documented that sexual recombination does not always occur when both mating types coexist in nature. In culture, ascomata form at the junction where A1 and A2 are paired. If the two mating types are incompatible or are sensitive to desiccation, they can withstand dry conditions much better than asexual spores, with the exception of chlamydospores.

www.Phytophtoradb.org

Known Diagnostic

Identification of *P. cinnamomi* has traditionally been performed via morphology and identification of distinct culture characteristics. A more recent approach to diagnosis is through the use of molecular methods. Molecular methods have the advantage of being able to detect and confirm the presence of *P. cinnamomi* with absolute certainty. Cultures of *PDA* are typically the root or conidia type; however, there are exceptions. Phytophthora with this approach have led to a wide variety of other identification schemes, including computer programs, which can identify the pathogen based on morphological features. A common method for diagnosis is to place a small amount of infected soil in a petri dish and allow it to dry. Once dry, the soil is placed in a petri dish containing 1 g of sterilized sand and 1 ml of water. After 24 hours, the soil is transferred onto the surface of PDA/4% V8 selects agar plates at a rate of approximately 0.1 ml of suspension per plate, then allowed to stand for 10 minutes. The inoculated plates are then incubated at 25°C for 3 days. After 3 days, the diameter of the colony is measured. In 1 mg of imiprotin, 250 mg of propiconazole, and 125 mg of triadimenolol. After a three day incubation period at room temperature, the diameter of the colony is measured again. The difference between the two measurements is the amount to be examined for colonies. Other various methods have been developed to test *P. cinnamomi* species from the soil by flooding, floatation or other host test in a water bath suspension. A suspension of the host in a plastic dish is kept then dried and flooded with water. The water is then removed, blotted on a paper towel, and placed on PDA/4% V8.

Control Strategies

Control efforts because of the pathogen's wide host range and ability to survive in symphatric or tolerant plants. Symphatric plants are a major source of spread to previously clean areas, which is major factor for field infections. Preventive measures include the avoidance of spreading the pathogen to new areas by not moving infected soil from one field to another. nursery through the use of clean seed and clean stock as well as utilizing well-drained sandy soils with a low pH. Control in propagation is best achieved by using clean propagation material, such as clean soil, clean containers, clean tools, and stakes. Using gravel beds on which to place pots. Disease severity can be reduced in planted nurseries by planting in raised beds. Fertilizers and lime can be used from combating the pathogen and promoting rapid drainage, the practice though may be effective, but it does not work. Chemical controls that may be present in the market will

Acknowledgments

Nominalistic information was provided by the Subramanyam Botany, and Mycology Laboratory in USDA-ARS. This specific page was written by Kelly Cross and Matt Greenway (NC State University), with picture contributions from Kelly Cross, Matt Greenway, Gary Grand, & Mike Bannan (NC State University), and Linda Hansen (Michigan University).

Notes

References

Aryalotta, J.P., Cross, K., and Guest, D.J. 2009. Suppression of *Phytophthora cinnamomi* in potting mixes amended with uncomposted and composted animal manures. *Phytopathology* 99:775-782.

Berren, D.M. and Grand, J.P. 2006. Incidence of *Phytophthora* root rot of Pepper in North Carolina. *Plant Disease* 90:1044-1046. doi:10.1094/PDIS-90-10-1044-A.

Chen, Y. and Grand, J.P. 2008. Root rot of pepper caused by *Phytophthora cinnamomi*. *Plant Disease* 92:1044-1046. doi:10.1094/PDIS-92-10-1044-A.

www.Phytophtoradb.org

Phytophtora Database - Windows Internet Explorer

Phytophtora DATABASE

Host : List

There are 179 hosts:

Common Name	Species Name	Wikipedia	Comment	Function
elder	[wiki]	[wiki]	[Modify]	[edit]
elafine	[wiki]	[wiki]	[Modify]	[edit]
almond	[wiki]	[wiki]	[Modify]	[edit]
ampalaya	[wiki]	[wiki]	[Modify]	[edit]
apple	[wiki]	[wiki]	[Modify]	[edit]
apicot	[wiki]	[wiki]	[Modify]	[edit]
esparagus	[wiki]	[wiki]	[Modify]	[edit]
ash	[wiki]	[wiki]	[Modify]	[edit]
avocado	[wiki]	[wiki]	[Modify]	[edit]
azalea	[wiki]	[wiki]	[Modify]	[edit]
baobab	[wiki]	[wiki]	[Modify]	[edit]
banana	[wiki]	[wiki]	[Modify]	[edit]
bamboo	[wiki]	[wiki]	[Modify]	[edit]
Bay Laurel	[wiki]	[wiki]	[Modify]	[edit]
bean	[wiki]	[wiki]	[Modify]	[edit]
beech	[wiki]	[wiki]	[Modify]	[edit]
beet	[wiki]	[wiki]	[Modify]	[edit]
begonias	[wiki]	[wiki]	[Modify]	[edit]

www.Phytophtoradb.org

Arbutus

From Wikipedia, the free encyclopedia

Arbutus (disambiguation)

Arbutus is a genus of flowering plants in the family Ericaceae, native to warm temperate regions of the Mediterranean, western Europe, and North America.

North American members of the genus are called Madrones, from the Spanish name madroño. The name "madroño" refers to the tree's resemblance of the hub of a strawberry; some species are sometimes referred to simply as the "Arbutus". Curiously, the name "Madrone" is used south of the Siskiyou Mountains of southern Oregon/Northern California and the name "Madrone" is used north of the Siskiyou Mountains according to the "Sunset Western Garden Book". North of the Canadian border, the name "Arbutus" is commonly used.^[1] All refer to the same tree, Arbutus menziesii, native to the Pacific Northwest and Northern California.

They are evergreen trees, shrubs growing to 5–25 m tall, with smooth or lenticular bark, usually arranged, due to broad terminal or lateral margins. The leaves are bell-shaped, 5–10 mm long, white or pink, and produced in racemes or corymb. The fruit is a rough-textured red or orange-red berry 1–2 cm diameter containing yellow flesh with numerous very small seeds; the fruit are edible but have minimal flavor and are not widely eaten.

A recent study which analyzed ribosomal DNA from *Arbutus* and related genera suggests that the Mediterranean Basin species of *Arbutus* are not very closely related to the North American species, and that the split between the two groups of species occurred at the Paleogene/Tertiary boundary.

Contents [show]

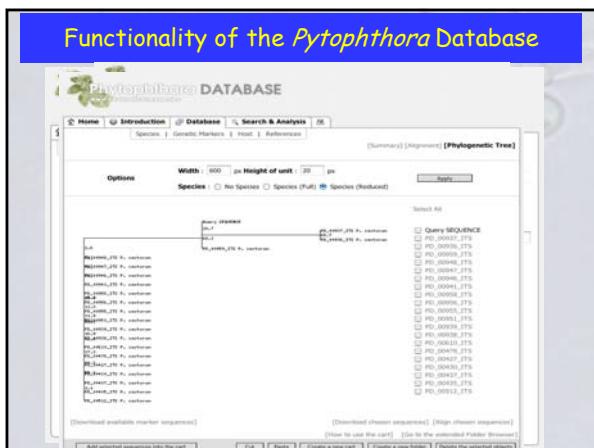
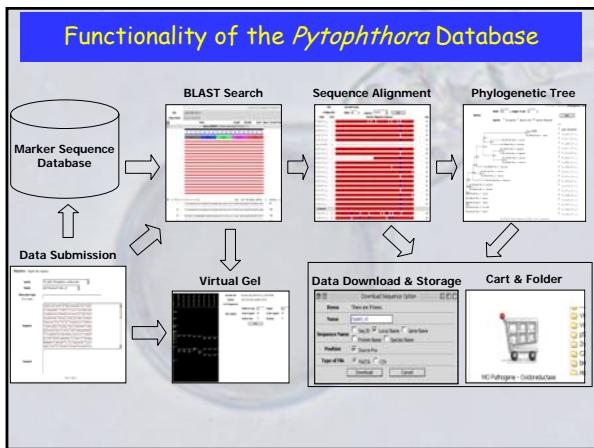
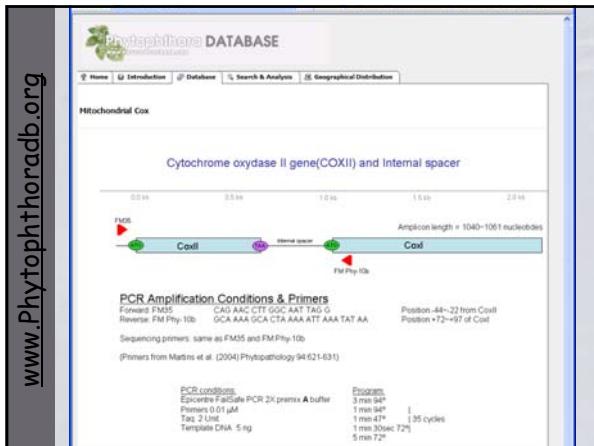
- 1 Species
- 1.1 Old World
- 1.2 New World
- 2 Uses and symbolism
- 3 Notes

Scientific classification

Kingdom: Plantae
Division: Magnoliophyta
Class: Magnoliopsida
Order: Ericales
Family: Ericaceae
Genus: *Arbutus*

Species

See text.



Global Phytophthora Network

Global Phytophthora Network

Phytophthora Database		Current Statistics	
(I) Phytophthora Database		Registered Users	>400
(II) Global Distribution		Countries	~50
		Species Covered	106
(III) Diagnostic Tools		Isolates Archived	2,356
		Genetic Marker Sequences	4,651
(IV) Analysis Tools		(V) Disease Management	
		VI Human Resource	
		International network Workshops Site visits Meetings Education Assistance & training in diagnosis Web-based resources	

Global Phytophthora Network

Global Phytophthora Network

Identification of ~2,600 isolates from multiple labs by sequencing the ITS region:

- 1. PA Dept. of Agriculture (~700)
- 2. UC-Riverside (~1,000)
- 3. Clemson University (~300)
- 4. University of Maryland (~200)
- 5. Chongbook National University in Korea (~150)
- 6. NC State (~100)
- 7. Ohio State University (~100)
- 8. Inst. for Plant Protection in Germany (~50)

-Assistance/training in diagnosis
-Web-based resources

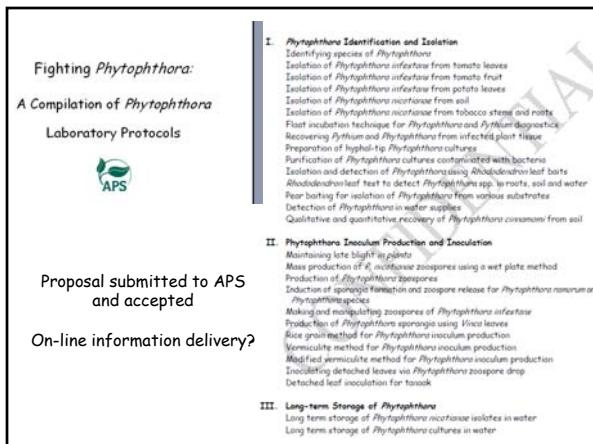
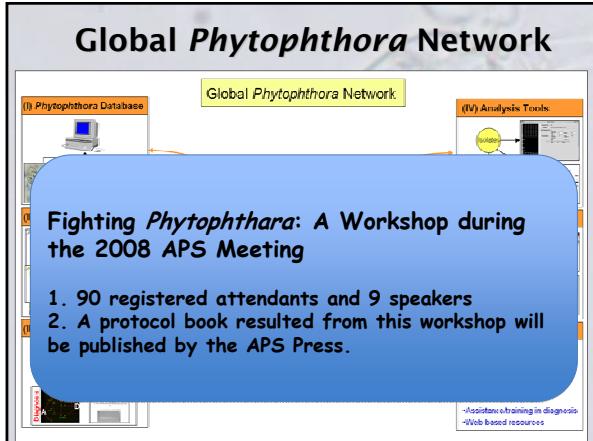
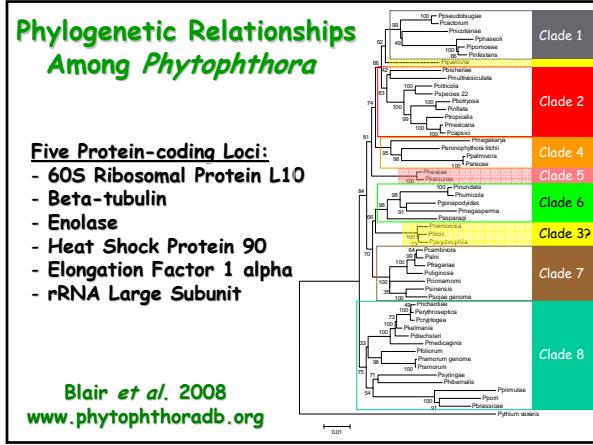
Global Phytophthora Network

Global Phytophthora Network

In Depth Phylogenetic Analysis & New Species Description:

- 1. Analysis of *P. capsici* and *P. cinnamomi* species complexes using 7 nuclear and 4 mitochondrial loci
- 2. New species description in collaboration with Yilmaz Balci (UMD), Mike Coffey (UC-Riverside), and Seong H. Kim (PDA)

-Assistance/training in diagnosis
-Web-based resources



Molecular Technology for Phytophthora identification and Development of Diagnostic Markers

Phytophthora DATABASE
www.phytophthora.org

Home | Introduction | Database | Search & Analysis | Refs. & News

Molecular Techniques for Isolate Identification and Development of Diagnostic Markers [Edit]

There are a number of molecular approaches available for identification of isolates to a species level, the selection of which one to use will depend on the type of analysis that is needed. If the isolate has been cultured the most accurate method for identification is DNA sequence analysis, but if it is not feasible to use this approach there are several gel based techniques that should work as well. For diagnosis from infected plant tissue there are PCR markers that are specific for detection of a Phytophthora species or group of species at the species or subspecies level. Several approaches are also available for following subpopulations of a particular species. While not fully developed at this point in time, work is in progress on the development of **microarrays** for identification of *Phytophthora* to a species level.

1. DNA sequencing
2. Gel based identification of species
3. Species specific diagnostic markers
4. Species specific subpopulation markers
5. Identification of subpopulations
5.1. Sequence analysis
5.2. Sequence Specific Polymerase Chain Reaction (SSPCR)
5.3. RFLP analysis
5.4. RAPDs and AFLP analysis
5.5. Mitochondrial haplotypes
5.6. Macro/ micro-arrays

Species-Specific Diagnostic Markers

Phytophthora DATABASE
www.phytophthora.org

Home | Introduction | Database | Search & Analysis | Refs. & News

Species specific diagnostic markers [Edit]

When the objective is to determine if a particular species is present in a plant sample, species-specific diagnostic markers are needed. These markers can be used to detect the presence of a particular species in a plant sample by either sequencing or amplifying a target region of the genome. A variety of sequences have been used for development of these diagnostic markers for a number of species in the genus *Phytophthora*. One of the most common approaches is to use a PCR system for detection of multiple species. For additional details [including reference segments used for primer development], see:

- ITS region
- rRNA genes
- Internal Transcribed Spacers
- Cox spacer region

In addition to conventional and real-time PCR there are several other molecular techniques that have been reported to be useful in detection of *Phytophthora* species. One of the most common approaches is to use a sequence specific probe that has been annealed to the target site in the ITS region of *P. infestans*. The resulting 40 bp fragment was diagnostic for *P. infestans* and the related species *P. brevis* and *P. sojae*. The sensitivity of detection was approximately 10 ng of total DNA. The sensitivity of detection was increased to a level of detection of the assay was increased to 0.1 ng. The sensitivity of detection was also increased by using a primer pair external to the target sequence and amplifying a larger region in a nested PCR reaction. This technique is called nested PCR and is used by many researchers. Another approach is to use a nested PCR system for detection of multiple species. One of the advantages of this technique is it doesn't require sophisticated equipment. A thermal cycler is sufficient for the assay or nested PCR, however, the sensitivity of detection is less for PCR (10 ng target DNA).

Considerations when developing species specific PCR diagnostic assays
 Standardized target size for PCR diagnostic assays
 Tardieu, M., Barker, N. 2007. Fungal, viral, arbovirus-specific methods for improved molecular detection of Phytophthora in the field. Applied and environmental microbiology. 73:4040-4047

Fitt, T., Truett, E., Hutchinson, S.L., Scott, and M.M. Caruso 2002 Use of ligase chain reaction for enhanced detection of Phytophthora infestans. Canadian Journal of Plant Pathology 24:296-305

Detailed Description of Diagnostic Markers

Phytophthora DATABASE
www.phytophthora.org

Home | Introduction | Database | Search & Analysis | Refs. & News

Species specific diagnostic markers [Edit]

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Fitt, T., Truett, E., Hutchinson, S.L., Scott, and M.M. Caruso 2002 Use of ligase chain reaction for enhanced detection of Phytophthora infestans. Canadian Journal of Plant Pathology 24:296-305

Sequence Alignment Files for Downloading

The screenshot shows a web-based database interface for Phytophthora. At the top, there's a blue header bar with the title "Sequence Alignment Files for Downloading". Below this is a search bar and a navigation menu with links like "Home", "Introduction", "Database", "Search & Analysis", "Tools & Resources", and "Help". The main content area has a sub-header "Species specific diagnostic markers". It contains several sections of text and diagrams related to PCR diagnostics. A prominent feature is a "Sequence Alignment" window with a green "Download" button. The bottom of the page includes a copyright notice and a reference to a paper by Tarran et al. (2007).

Reference Management System

This screenshot shows the same Phytophthora Database interface as the previous one, but the main content area is now focused on a "Reference Management System". It displays a "Reference Browser" section with a "Detail Information of the paper" panel. This panel contains fields for "Title", "Abstract", "Authors", "Journal", "Affiliation", "Institute", and "Creator". The "Abstract" field describes a method for improved molecular detection of *Phytophthora ramorum*. The "Journal" field lists "Tarran, J., Berlin, I., Beaman, S., 2007. Faster, simpler, more specific methods for improved molecular detection of Phytophthora ramorum in the field. Applied and environmental microbiology, 73(466-467)". The "Affiliation" field lists "Centre for Science Laboratory, Sand Hutton, York YO4, United Kingdom, J. tarran@csil.gov.uk". The "Institute" field lists "University of York NRI". The "Creator" field lists "J. Tarran (Sergiou Park)".

Geographic Information Systems Tools

This screenshot shows the "Geographic Information Systems Tools" section of the Phytophthora Database. It features a map of the world titled "Geographical distribution" with various regions highlighted in different colors. Below the map is a "About this map" section containing text about the map's source and a "Map credits" section listing "USDA ARS Agricultural Research Service" and "USGS". There's also a "Map scale" section with options for "1:100,000", "1:1,000,000", and "1:10,000,000". At the bottom of the page is a copyright notice and a reference to a paper by Tarran et al. (2007).

