

- USDA-APHIS-PPQ-Center for Plant Health Science & Technology (CPHST Beltsville Laboratory) is almost ready to officially release the “IDphy: Molecular and Morphological Identification of *Phytophthora* based on the Types”. **This is the first online resource of this nature implemented for plant pathogens ([link](#)).**
- The Molecular tool box of *Phytophthora* with seven genes from a range of well authenticated species representatives including the Ex-types has now close to 1000 sequences ([link](#)) .
- Work with 3rd Generation High-Throughput Sequencing (3G-HTS) via MinION for identification for *Phytophthora* is in progress.
- We are working with Whole Genome Sequencing (APS 2019 Poster 690-P2. Srivastava et al. WGS of the Ex-type of *P. ramorum*), metabarcoding and metagenomics (the last in collaboration with Oklahoma State University).

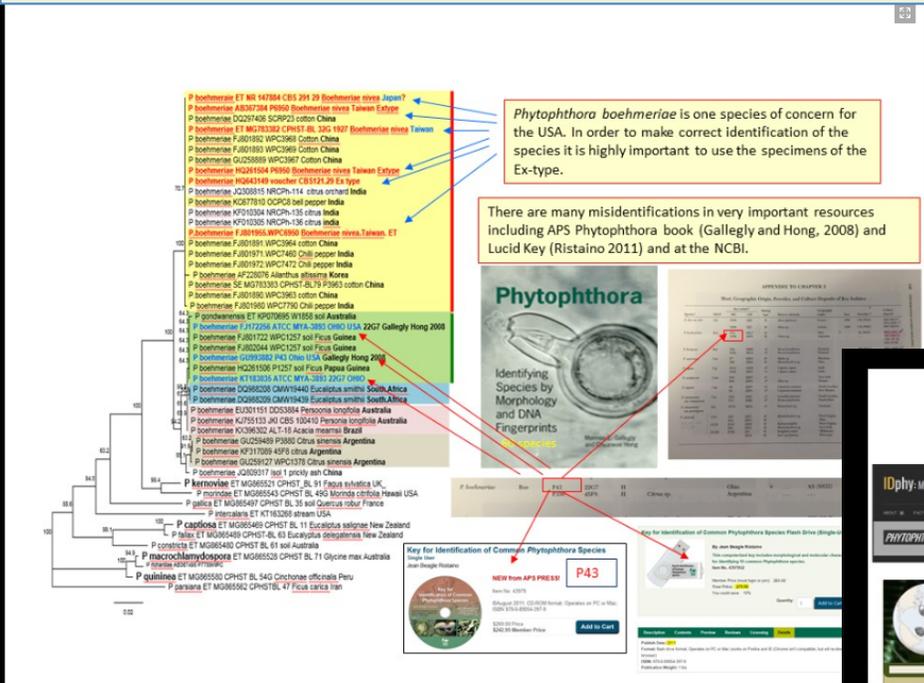


The case of *P. boehmeriae* species of concern for the USA and other countries. This species is in Asia

Sequencing identification at NCBI and other resources is challenging due to the proliferation of misidentified specimens. Proliferation of incorrect citations in online and printed resources is of worldwide concern

The distribution of *P. boehmeriae* is in Asia.

This species is incorrectly cited in several online resources that indicate hosts and distributions in Latin America (Brazil, Argentina), Australia, New Guinea, South Africa, Ohio (USA).



Phytophthora species in Clade 10 (ITS rDNA). Notice the position of the cluster of the "sensu stricto" of *P. boehmeriae*, in relation of the position of *P. gondwanensis* and other related clusters. Blue arrows point the position of sequences from the ex-type and red arrows show the position of misidentifications.

Phytophthora species in Clade 10 (ITS rDNA). Notice the position of the cluster of the "sensu stricto" of *P. boehmeriae*, in relation of the position of *P. gondwanensis* and other related clusters. Blue arrows point the position of sequences from the the ex-type and red arrows show the position of misidentifications.

Information for the species in other databases:

- Phytophthora DATABASE: *Phytophthora boehmeriae*
- Q-bank: *Phytophthora boehmeriae*
- SMML USDA-ARS: *Phytophthora boehmeriae*
- EPPO Global Database: *Phytophthora boehmeriae*
- Forest Phytophthoras of the world: *Phytophthora boehmeriae* (Incorrectly cites the occurrence of the species in different parts of the world).
- CABI Invasive Species Compendium: *Phytophthora boehmeriae*
- Encyclopedia of Life (EOL): *Phytophthora boehmeriae*
- Index Fungorum (IF): *Phytophthora boehmeriae*
- Plantwise Knowledge Bank: *Phytophthora boehmeriae*

Abstract

Distribution of *Phytophthora nicotianae* and *P. boehmeriae* in Brazilian plantations of black wattle.

To determine the distribution of *P. nicotianae* and *P. boehmeriae* in Brazilian black wattle (*Acacia mearnsii*) plantations, a survey was conducted in 23 plantations located in 9 municipalities of Rio Grande do Sul and 2 experimental areas located in 2 municipalities of Paraná. *P. nicotianae*, which...

Author(s): Santos, Á. F.; Luz, E. D. M. N.
 Publisher: Sociedade Brasileira de Fitopatologia, Brasília, Brazil
 Citation: Fitopatologia Brasileira, 2006, 31, 4, pp 398-400

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The distribution of *P. boehmeriae* is in Asia. *Phytophthora boehmeriae* is incorrectly cited in several online sources which indicate hosts and distributions in Latin America (Brazil, Argentina), Australia, New Guinea, South Africa, Ohio (USA).

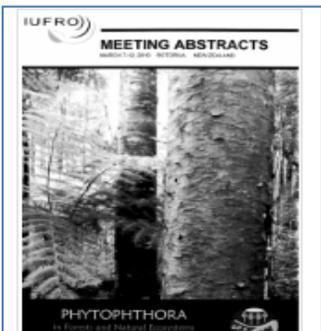


The Taxonomy of *Phytophthora*: What is done from our CPHST BL?



Presented the importance of the Ex-types in different national and international forums. Including IUFRO Phytophthora

<http://forestphytophthoras.org/proceedings>



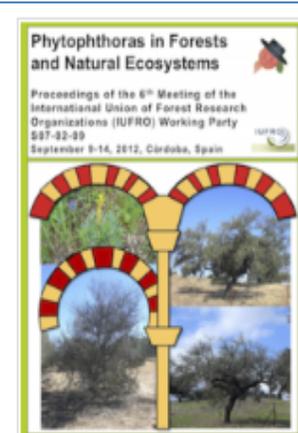
March 7-12, 2010. Auckland and Rotorua, New Zealand.

Refined systematics (taxonomy, nomenclature and phylogenetics) of *Phytophthora* for more accurate morphological-molecular identification: The importance of types and authenticated specimens.

Z. Gloria Abad¹, and Michael C. Coffey²

- 2010**
- Link to New Zealand Journal of Forestry Science Supplement
 - Link to conference abstracts

March 7-12, 2010. Auckland and Rotorua, New Zealand. Pdf icon link will download the book of abstracts, the NZJFS link has downloads for the published articles.



September 9-14, 2012. Cordoba, Spain.

Morphological-Molecular ID Tools of *Phytophthora*: Lucid & Tabular Keys and Sequencing Analysis

Z.G. Abad¹, Y. Balci², M.D. Coffey³, S. Kang⁴

2012
September 9-14, 2012

Córdoba, Spain



10th - 14th November 2014. Esquel, Chubut. Patagonia Argentina

The Taxonomy of *Phytophthora*:

What is done and what is needed for the correct identification and diagnostics of species in the Genus

Z. G. Abad

10th - 14th November 2014
2014
Esquel, Chubut. Patagonia Argentina

Sutton, W., Reeser, P.W., and Hansen, E.M., tech coords. 2015. 197 P.



United States Department of Agriculture

The Taxonomy of *Phytophthora*: What is done from our CPHST BL?

Presented the importance of the Ex-types in different national and international forums. Including “Oomycetes International Workshops” [Link: https://www.isppweb.org/smc_10.asp](https://www.isppweb.org/smc_10.asp)

International Society for Plant Pathology

Promoting World-Wide Plant Health and Food Security

Home Congresses Commission on Global Food Security Newsletter Archive Subject Matter Committees Resources Administration

ISPP Subject Matter Committee: Oomycetes

Chair: Dr Gloria Abad

Address: Oomycetes-Fungi Leader, USDA-APHIS-PPQ-Center for Plant Health Science & T

Phone: 301 313 9340

Fax:

Mobile:

Email: gloria.abad@aphis.usda.gov

Coming Events

Past Events

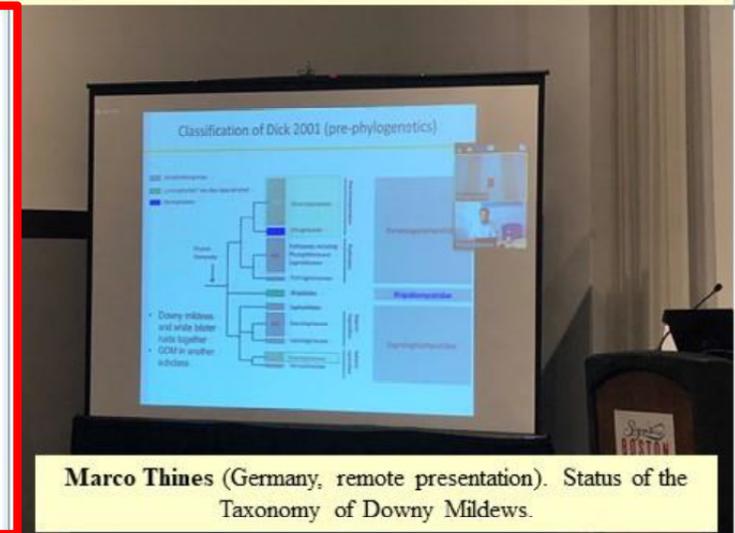
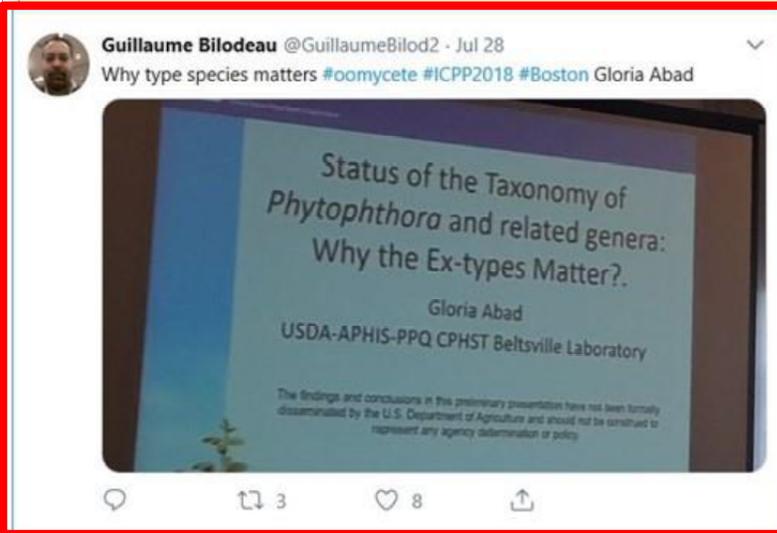
- 1st International Oomycetes Workshop 23-27 July, 2004
- 2nd International Oomycetes Workshop May 21-26 2006
- 2nd International Oomycetes Workshop Photos
- 3rd International Oomycetes Workshop. ICPP 2008
- 3rd International Oomycetes Workshop. 2008 photos.
- 3rd International Oomycetes Workshop. ICPP 2008 at
- 3rd International Oomycetes Workshop Posters
- 3rd International Oomycetes Workshop Presentations
- 3rd International Oomycetes Workshop Presentations
- 3rd International Oomycetes Workshop Visitor Speak
- 3rd International Oomycetes Workshop ISPP 39-2 20
- 4th Int Oomycetes Workshop. Maryland USA 2012.pd
- 6th International Oomycetes Workshop Boston 2018

Members of the Oomycetes Committee 21st March 2018

6th International Oomycetes Workshop – ICPP 2018

108 participants from 25 countries, including 28 speakers

Session #1: Taxonomy and Nomenclature: Past, present, and future. Moderator: Hai Nguyen (Canada)



Marco Thines (Germany, remote presentation). Status of the Taxonomy of Downy Mildews.



United States Department of Agriculture

The Taxonomy of *Phytophthora*: What is done from our CPHST BL?

Presented the importance of the Ex-types in different national and international forums. Including “Oomycetes International Workshops” [Link: https://www.isppweb.org/smc_10.asp](https://www.isppweb.org/smc_10.asp)

ICPP 2018 - Sixth International Oomycetes Workshop

Oomycetes in the era of Plant Health in A Global Economy

Boston, Massachusetts, USA - July 28, 2008

Workshop organized by the ISPP – SMC Oomycetes to celebrate the 50th anniversary of ISPP



What is needed for correct species identification?

IDphy: Molecular and Morphological identification of *Phytophthora* based on the types

IN BETA TESTING 7.31.19

IDphy: Molecular and morphological identification of *Phytophthora* based on the types

Molecular and morphological identification of *Phytophthora* species based on the types and other well-authenticated specimens

IDphy was developed to facilitate accurate and efficient identification of *Phytophthora* to species, using type specimens from the original descriptions for reference wherever possible. IDphy emphasizes species of high economic impact and species of regulatory concern for the U.S. IDphy includes molecular and morphological tools to aid in identification of the 161 culturable species of *Phytophthora* described as of May 2018.

- ★ **Molecular sequences and protocols**
Tools to support molecular identification include sequence vouchers for ITS rDNA and COI, as well as protocols and SOPs for DNA extraction, PCR, electrophoresis gels, and sequence-based identification. All voucher sequences in the fact sheets are from the types or from selected well-authenticated specimens.
- ★ **Morphological keys and culturing protocols**
Morphology-based support includes an interactive Lucid key; a tabular key; over 160 fact sheets drawn from the type specimens whenever possible; an image gallery with over 1000 images of phylogenetic trees, morphological features, and disease symptoms; and protocols for isolation, growth, sporulation, and storage.

Section of FACT SHEET INDEX showing a portion of the list of the 161 culturable species in letter A alphabetical order.

USDA

Section of ABOUT THIS TOOL showing a portion of “Tool background and scope” and the list of different topics presented at this Section

IDphy: Molecular and morphological identification of *Phytophthora* based on the types

ABOUT THIS TOOL

- > Tool background and scope
- > What is *Phytophthora*?
- > History of *Phytophthora*
- > Authors
- > External links
- > Acknowledgments
- > Copyright, citation, and disclaimers

FACT SHEET INDEX

A B C D E F G H I K L M N O P Q R S T U < V X

A

- ▶ *Phytophthora acerina*
- ▶ *Phytophthora alni* subsp. *alni*
- ▶ *Phytophthora alni* subsp. *uniformis*
- ▶ *Phytophthora amarantii*
- ▶ *Phytophthora andina*
- ▶ *Phytophthora arenaria*
- ▶ *Phytophthora asparagi* (invalid)
- ▶ *Phytophthora austrocedri*
- ▶ *Phytophthora agathidicida*
- ▶ *Phytophthora alni* subsp. *multiformis*
- ▶ *Phytophthora alticola*
- ▶ *Phytophthora amnicola*
- ▶ *Phytophthora aquimorbida*
- ▶ *Phytophthora asiatica*
- ▶ *Phytophthora attenuata*

MOLECULAR IDENTIFICATION

MOLECULAR PROTOCOLS

In order to facilitate the correct identification of *Phytophthora* species, we provide standards of procedures (SOPs) for all steps involved with molecular identification using sequencing analysis of the ITS rDNA and COI barcoding regions. Each SOP is available as a downloadable PDF:

- ☐ SOP-PID-01.00 Molecular: Equipment, materials, and reagents for DNA extraction and PCR
- ☐ SOP-PID-02.00 Molecular: DNA extraction for *Phytophthora* species using Qiagen kit (suggested)
- ☐ SOP-PID-03.00 Molecular: Primers dilutions for PCR amplifications
- ☐ SOP-PID-04.00 Molecular: Gels, electrophoresis, and photos
- ☐ SOP-PID-05.03 Molecular: Identification of *Phytophthora* species with ITS rDNA and COI barcoding genes

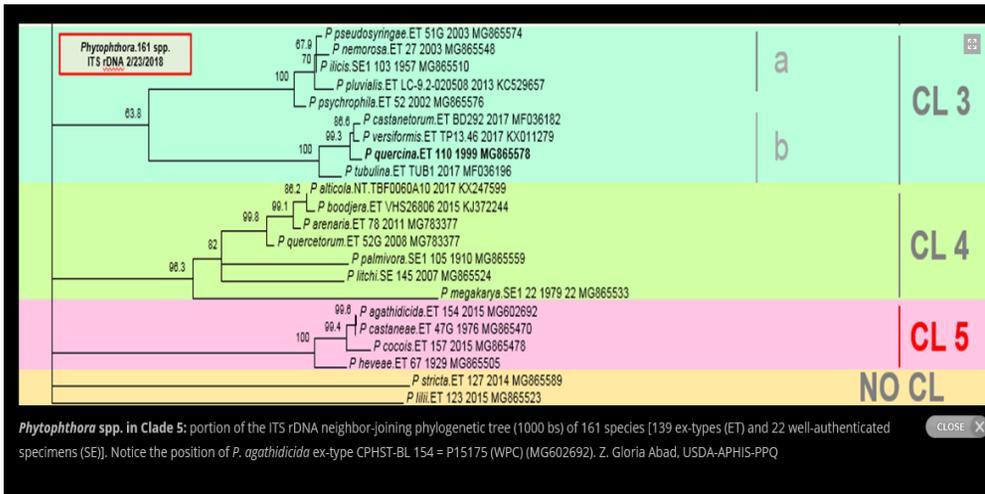
IDENTIFYING PHYTOPHTHORA UNKNOWN

For quick reference, the steps from page 6 of SOP-PID-05.02 are presented here. Follow these **important** steps to identify *Phytophthora* unknown species using vouchers of the ITS rDNA gene of 139 ex-types and 22 well-authenticated species:

Suggested citation: Abad, Z.G., Burgess T., Bienapfl J.C., Redford A.J., Coffey M., and Knight L. 2019. IDphy: Molecular and Morphological Identification of *Phytophthora* Based on the Types. USDA APHIS PPQ S&T Beltsville Lab, USDA APHIS PPQ S&T ITP, and Centre for *Phytophthora* Science and Management. <http://idtools.org/id/phytophthora/index.php> <date you accessed site>

IDphy: Molecular and Morphological identification of *Phytophthora* based on the types

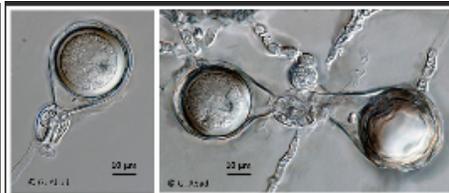
Some of the figures included in the Factsheet of *Phytophthora agathidicida*. **LEFT:** ITS rDNA partial phylogenetic tree showing species in Clades 3, 4 and 5. Marked in red is CL5 that contains *P. agathidicida*. Colonies and morphology of the Ex-type CPHST BL 154. **RIGHT:** Kauri dieback caused by *P. agathidicida* in New Zealand and action to control spread of the disease. Similar set of figures is included for each of the 161 species included in the resource.



taking action to prevent the spread of kauri dieback, caused by *Phytophthora agathidicida*; photo courtesy of Kauri Dieback Programme Partners, New Zealand



Phytophthora agathidicida (CPHST-BL 154) colonies of the ex-type grown for 7 days on (a) V8® Agar, (b) potato dextrose agar, and (c) malt extract agar; photo by Krysta Jennings and Leandra Knight, USDA-APHIS-PPQ



Phytophthora agathidicida (ex-type CPHST-BL 154) sexual phase formed on hemp seed agar: oogonia with wavy wall and tapered base, amphyginous antheridia, and plerotic, thick-walled oospores; photos by G. Abad, USDA-APHIS-PPQ

Kauri giant tree with symptoms of bleeding canker. The New Zealand expert holds in the left hand a piece of the cortex that will be replaced, in respect of indigenous Māori traditions. Kauri holds a very significant place in their creation mythology, with the iconic trees having their own names, for example Tāne-mahuta “the God of the Forest”. photo by Gloria Abad, USDA-APHIS-PPQ



Kauri dieback, caused by *Phytophthora agathidicida*, is specific to New Zealand kauri and can kill trees of all ages; photo courtesy of Kauri Dieback Programme Partners, New Zealand.



NCBI Resources How To Sign in to NCBI

Nucleotide Nucleotide Phytophthora AND CPHST AND BL Search

Create alert Advanced Help

Species Summary 20 per page Sort by Default order Send to Filters: Manage Filters

Protists (991)
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Molecule types
genomic DNA/RNA (991)
Customize ...

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INSDC (GenBank) (991)
Customize ...

Sequence Type
Nucleotide (991)

Genetic compartments
Mitochondrion (181)

Sequence length
Custom range...

Release date
Custom range...

Revision date
Custom range...

[Clear all](#)

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See [BI Bristle](#) in the Gene database
bl reference sequences

Items: 1 to 20 of 991

<< First < Prev Page 1 of 50 Next > Last >>

- [Phytophthora pseudolactucae strain Ex-type CPHST BL 118 internal transcribed spacer 1, partial sequence; 5.8S ribosomal RNA gene and internal transcribed spacer 2, complete sequence; and large subunit ribosomal RNA gene, partial sequence](#)
850 bp linear DNA
Accession: MG865573.1 GI: 1336651463
[Taxonomy](#)
[GenBank](#) [FASTA](#) [Graphics](#) [PopSet](#)
- [Phytophthora ramorum strain Ex-type CPHST BL 55G, whole genome shotgun sequencing project](#)
1,322 rc linear DNA
i This entry is the master record for a whole genome shotgun sequencing project and contains no sequence data.
Accession: RYEP00000000.1 GI: 1589400222
[Assembly](#) [BioProject](#) [BioSample](#) [Taxonomy](#)
[GenBank](#)
- [Phytophthora uliginosa strain Ex-Type CPHST BL 59 translation elongation factor 1-alpha \(EF1A\) gene, partial cds](#)
875 bp linear DNA
Accession: MH359083.1 GI: 1409190206
[Protein](#) [Taxonomy](#)
[GenBank](#) [FASTA](#) [Graphics](#) [PopSet](#)

Results by taxon

Top Organisms [Tree](#)

- Phytophthora meadii (22)
- Phytophthora palmivora (21)
- Phytophthora infestans (20)
- Phytophthora nicotianae (20)
- Phytophthora melonis (19)
- All other taxa (889)

More...

Find related data

Database:

Search details

("Phytophthora" [Organism]
OR Phytophthora [All
Fields]) AND CPHST [All
Fields] AND BL [All Fields]

[See more...](#)

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🔍 Phytophthora AND CPHST AND BL (991)

<https://www.ncbi.nlm.nih.gov/nucore/?term=Phytophthora+AND+CPHST+BL>

CPHST BL sequences of ITS rDNA form the Ex-types to be placed in RefSeq at NCBI

ResearchGate
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Project

Oomycetes ITS markers in RefSeq

Conrad L Schoch ·
 Barbara Robbertse ·
 Diana Haddad ·
 [Show all 6 collaborators](#)

Goal: The project will provide reference targeted loci to aid accurate assessment of species names in Oomycetes, focused on ITS barcode markers from type material. This will expand the existing framework for Fungi ITS RefSeq (PRJNA177353).

Curation actions:

- Define a list of Oomycete species and assess taxonomic accuracy
- Verify a complete set of type material data in NCBI Taxonomy
- Select and verify Nucleotide records for RefSeq inclusion
- Rely on feedback from expert collaborators

Updates 0 new 1

Recommendations 0 new 3

Followers 0 new 23

Reads 1 new 152

6 collaborators

- Conrad L Schoch [View profile](#)
- Barbara Robbertse [View profile](#)
- Diana Haddad [View profile](#)
- Pooja K Strope [View profile](#)
- Zoila Gloria Abad [View profile](#)
- Subodh Srivastava
Not yet on ResearchGate

Project log References (4) Questions

Project

Conrad L Schoch
added an **update**

NCBI project page

NCBI project page is here:

Accession: PRJNA362621 ID: 362621

<https://www.ncbi.nlm.nih.gov/bioproject/362621>

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1 Comment · 3 Recommendations · 96 Reads

...**Andrey M Yurkov**
il 52.66 · Leibniz Institut DSMZ - Deutsche Sammlung von Mikroorganismen und...

Great! Looking forward to see more RefSeq data.

Progress of the CPHST-BL in 3rd Generation High-Throughput Sequencing (3G HTS)

WGS via MinION 3rd generation HTS data generation

P. ramorum CPHST #55G P10103 ET

- Date of Extraction: 8/17/18
Run 7
- Volume:
 - Qubit: 104 ng/ul
 - Nano-drop conc: 47.2 ng/ul
 - 260/280: 1.94
 - 260/230: 1.86
 - DIN: 6.8
 - Max Molecular Weight: 57,801

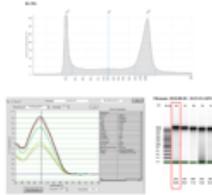


Fig 3. High quality (HQ) DNA obtained from pure culture isolate of the Ex-type of *Phytophthora ramorum* using Qiagen DNeasy Mericon Kit. Observe the DNA integrity number (DIN = 6.8) and the ratios for the 260/280 and 260/230 obtained with Nano-drop.

Table 1: Genome assembly statistics of *Phytophthora ramorum* strain CPHST-BL 55G (duplicate of Ex-type CBS 10553) via MinION long-reads using Canu assembler.

Attribute	Numbers	Attribute	Numbers
Total generated reads	2,676,135	N50	97267
Quality reads >1000 (bases)	625,764	Max	588354
Mean read length	2673	Sum	52430000
Average coverage	42X	CorMinCoverage	4
Total assembled contigs (>1000)	1322	Option (CorMhap Sensitivity)	High
L50	147	Contigs >100 kb	139
Min	1006	GC %	54.7

CBS: Culture collection of the Cebrtaalbureau voor Schimmelcultures, Fungal Biodiversity Centre, Utech, The Netherlands. CPHST-BL: Center of Plant Health Science and technology - Beltsville Laboratory, United States Department of Agriculture.

We have successfully generated a WGS of the Ex-type of *P. ramorum* CPHST BL #55G (= CBS 101553).

We have developed new ITS-targeting primers that were designed to work for metabarcoding starting from environmental DNA (e-DNA) samples.

Metabarcoding via MinION 3rd generation HTS analysis

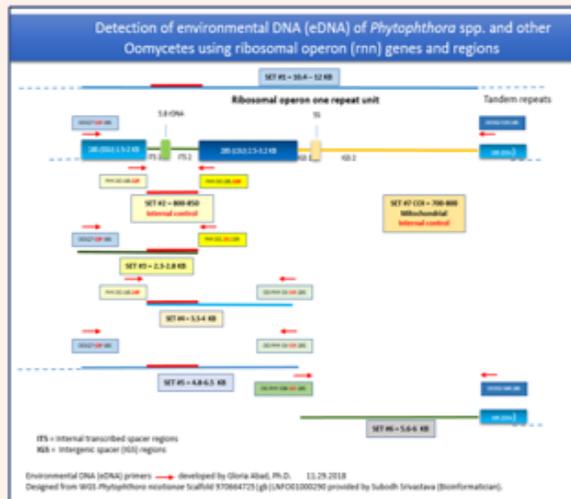


Fig 4. Primers developed from the ribosomal operon (12kb, 6kb, 4kb, 2.5kb, 800bp and 750bp). The best primers amplified the ITS rDNA (800-850bp).

Fig 5. 1st generation (Sanger) didn't resolve barcodings BC08 and BC09. These combinations were resolved by MinION reads.



Fig 6. Analysis results of two and more pathogens infections via MinION 3rd generation sequencing



United States Department of Agriculture

Thank you!

