Community ecology of foliar fungi and oomycetes of *Pseudotsuga menziesii*

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MS Defense

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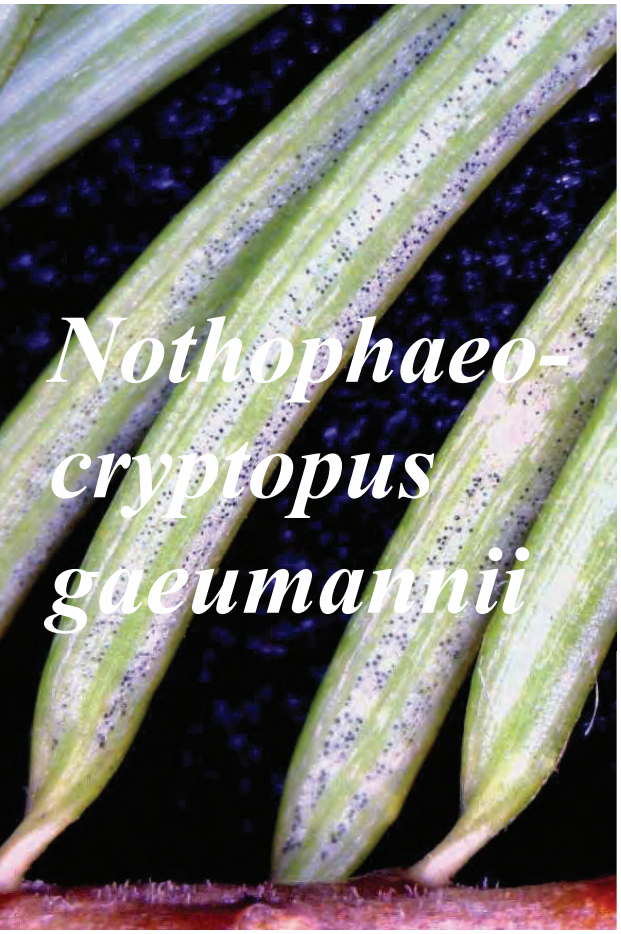
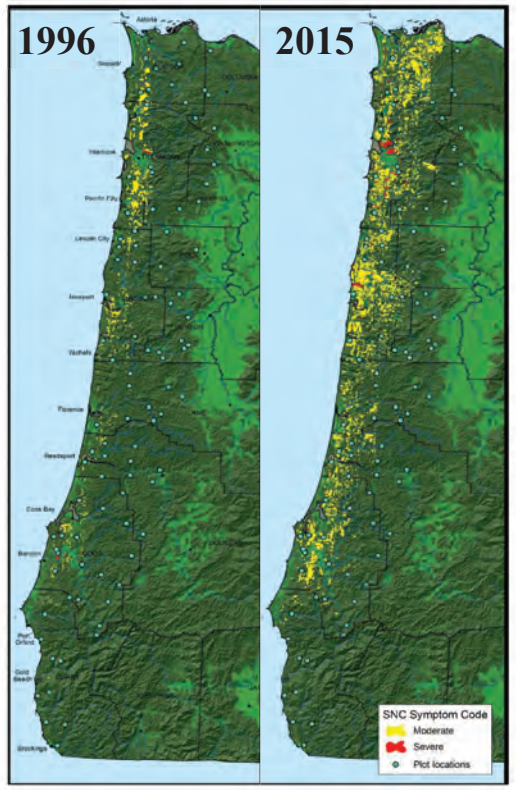
Advised by Jared LeBoldus and Andy Jones



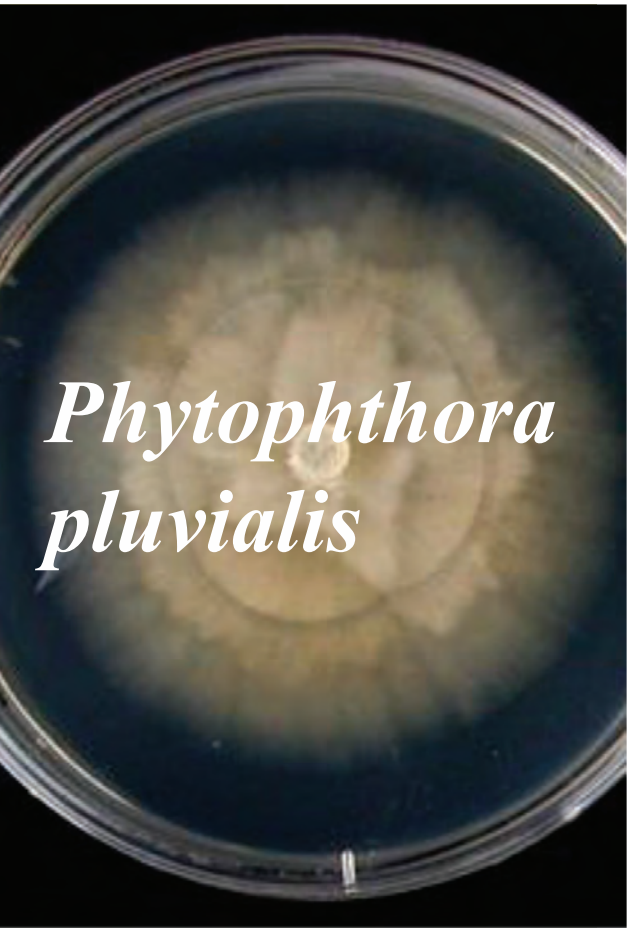
OSU SNC Cooperative

Fungal endophyte/latent pathogen

- **Ubiquitous, obligate, specific to *Pseudotsuga***
- **Causal agent of Swiss needle cast (SNC)**
  - Climate change (Manter et al. 2005)
  - Management (Stone et al. 2008)
  - Climate transfer distance (offsite planting) (Wilhelmi et al. 2017)



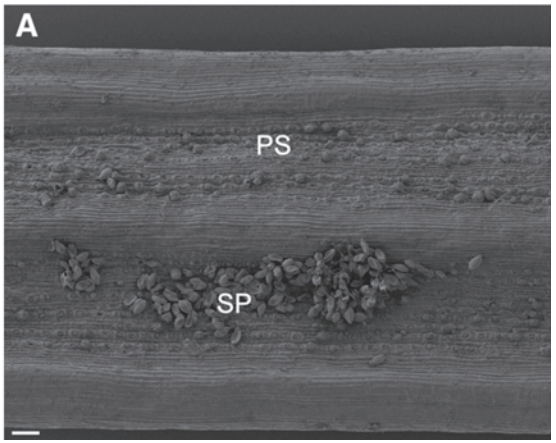
Adapted from Ritóková et al. 2016



Reeser et al. 2013

Only foliar oomycete described in *P. menziesii*

- **Cryptic symptoms**
- **Episodic, geographically isolated needle casting events**
- **Inversely abundant to *N. gaeumannii*** (Gómez-Gallego et al. 2019)



Gómez-Gallego et al. 2019

Objectives

1) Describe community dynamics of foliar fungi and oomycetes of *P. menziesii* on the PNW coast

2) Characterize microbial communities in response to effects of host location and genetic lineage

3) Model fungal diversity in response to climate transfer distance and oomycete diversity

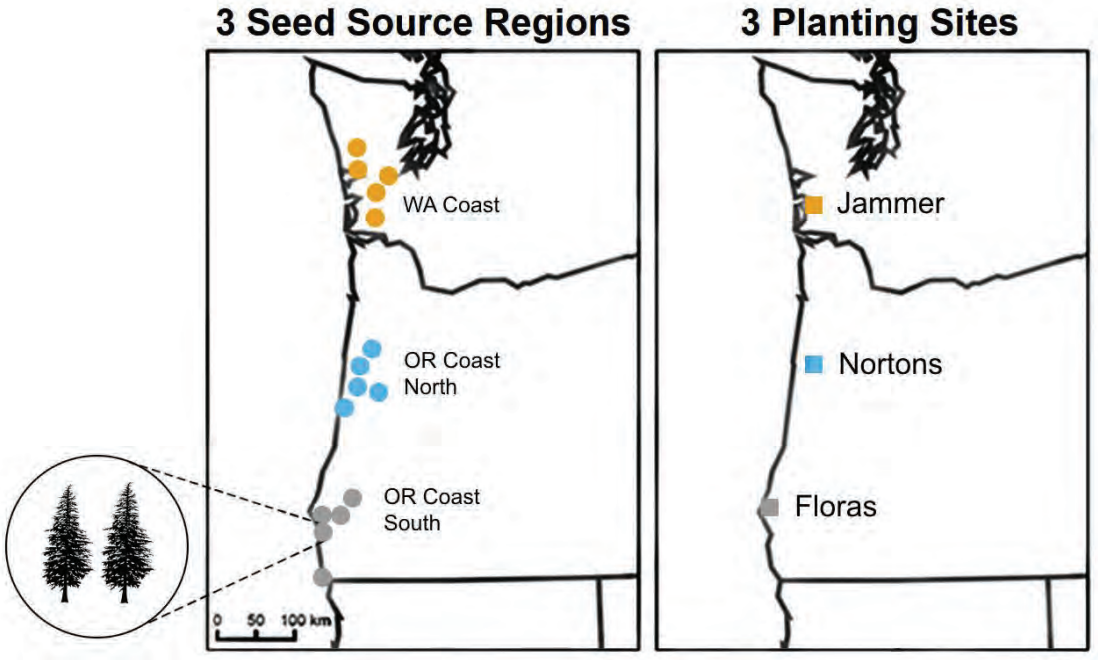
4) Investigate associations between *N. gaeumannii* and oomycetes

Methods

- **Sample needles from trees in reciprocal transplant provenance study (SSMT)**
- **Culture**
  - Isolate and sequence oomycetes from needles
- **Next-generation sequencing (NGS)**
  - Sequence all fungal and oomycete DNA extracted from needles
- **Statistical analyses**
  - Analyses of community dissimilarity
  - Generalized linear mixed modeling (GLMM)

Study Sites

USFS Douglas-Fir Seed Source Movement Trial (SSMT)

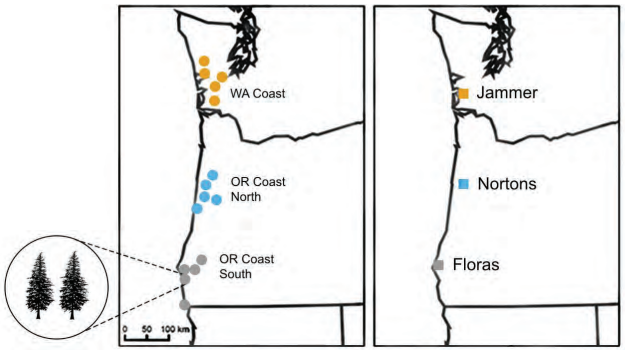


Adapted from Wilhelmi et al. 2017

Study Sites

Block 4		Block 3	
43	42	31	30
44	41	32	29
45 WACST	40 ORCSTS	33 WACST	28 ORCSTS
46	39 ORCSTN	34 ORCSTN	27
47	38	35	26
48	37	36	25
19	18	7	6
20 WACST	17	8	5
21	16	9 ORCSTS	4 WACST
22 ORCSTN	15	10	3 ORCSTN
23 ORCSTS	14	11	2
24	13	12	1
Block 2		**Block 1**	

7201	8027	7202	8028
8027	8026	7X5	7206
7201	8542	8028	8541
8026	7206	7205	8025

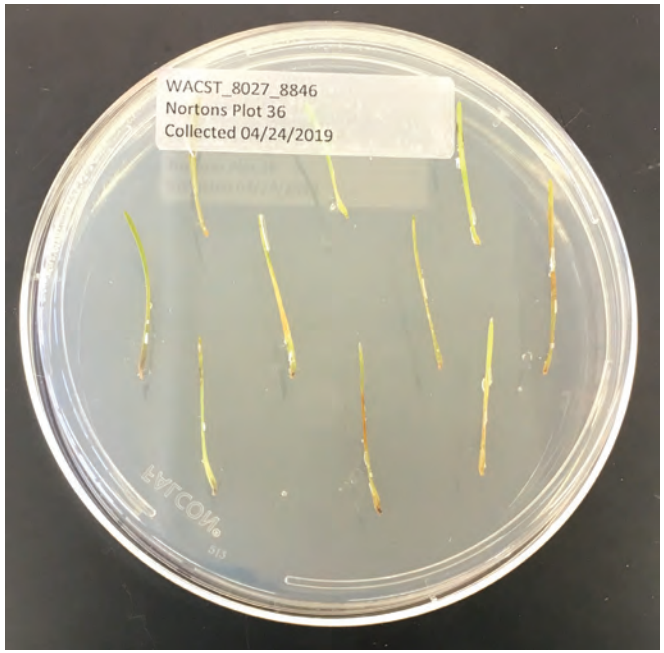


n = 338

Sample Collection

Sample Collection

Plate 10 needles

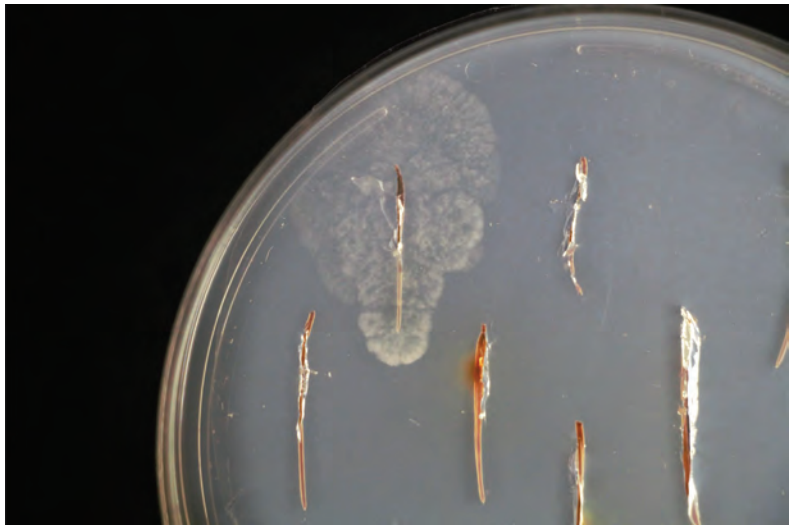


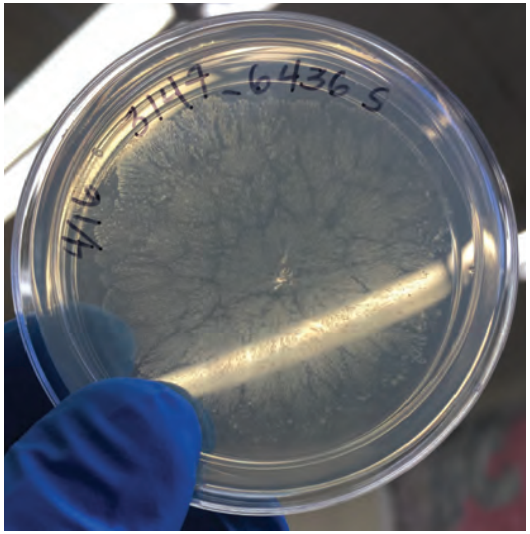
Sequence 10 needles

Culture

18 isolates

Nortons: 16 Jammer: 2 Floras: 0





Culture

12 isolates

Nortons: 11 Jammer: 1 Floras: 0

ITS

- ITS5 & ITS4 (White et al. 1990)

COI

- OomCoxI-Levup & OomCoxI-Levlo (Robideau et al. 2011)

Rps10

- Prv9r-M & Prv9f-M (Foster 2020)

NGS

Library prep and sequencing

- **2-stage PCR**
  - Fungi: **ITS2**
  - Oomycetes: ***Rps10***
- **Illumina MiSeq 3000**
  - 300-bp paired end reads
  - Fungi and oomycetes sequenced on same lane

Illumin a

NGS

Fungi

- n = 246
- OTUs = 105

Oomycetes

- n = 181
- OTUs = 55

All microbes

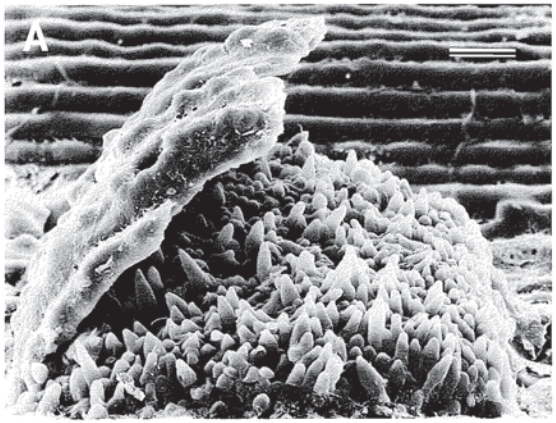
- n = 282
- OTUs = 157
- Rarefied to 1000 min reads

	OTU.1	OTU.2	OTU.3	OTU.5
F12-3180-1757.f	4.009315	2.432123	3.163402	2.50086
F12-6059-2776.f	3.902164	3.292552	3.396323	0.00000
F12-6059-7502.f	3.965152	3.228200	2.447291	0.00000
F12-8552-4829.f	4.004358	0.000000	3.008734	0.00000
F12-8552-9359.f	3.780196	3.568014	2.988478	0.00000
F14-3147-1661.f	3.889971	0.000000	2.458972	0.00000

1) Describe community dynamics of foliar fungi and oomycetes of *P. menziesii* on the PNW coast

Fungi (105 OTUs)

OTU	Taxon
OTU 1	*Rhabdocline parkeri*
OTU 2	*Rhizosphaera* sp.
OTU 3	*Nothophaeocryptopus gaeumannii*
OTU 4	*Rhizosphaera* sp.
OTU 5	*Curvibasidium cygneicollum*
OTU 7	*Cladosporium* sp.
OTU 8	*Penicillium penicilliodes*
OTU 9	*Hormonema macrosporum*
OTU 10	*Curvibasidium cygneicollum*
OTU 12	Didymellaceae



Sherwood-Pike et al. 1986

OSU SNC Cooperative

1) Describe community dynamics of foliar fungi and oomycetes of *P. menziesii* on the PNW coast

Oomycetes (55 OTUs)

OTU	Taxon
OTU 1	*Phytophthora cacuminis*
OTU 4	Pythiaceae
OTU 7	Oomycete
OTU 8	Oomycete
OTU 11	Oomycete
OTU 13	Oomycete
OTU 16	Oomycete
OTU 18	Oomycete
OTU 22	Pythiaceae
OTU 26	Oomycete

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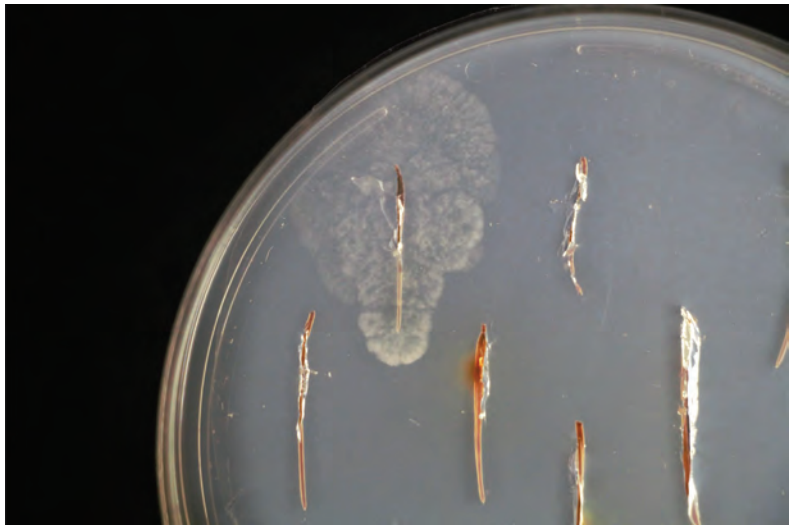

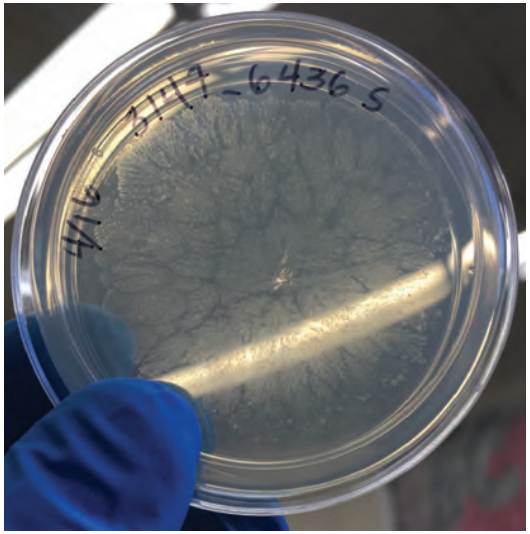

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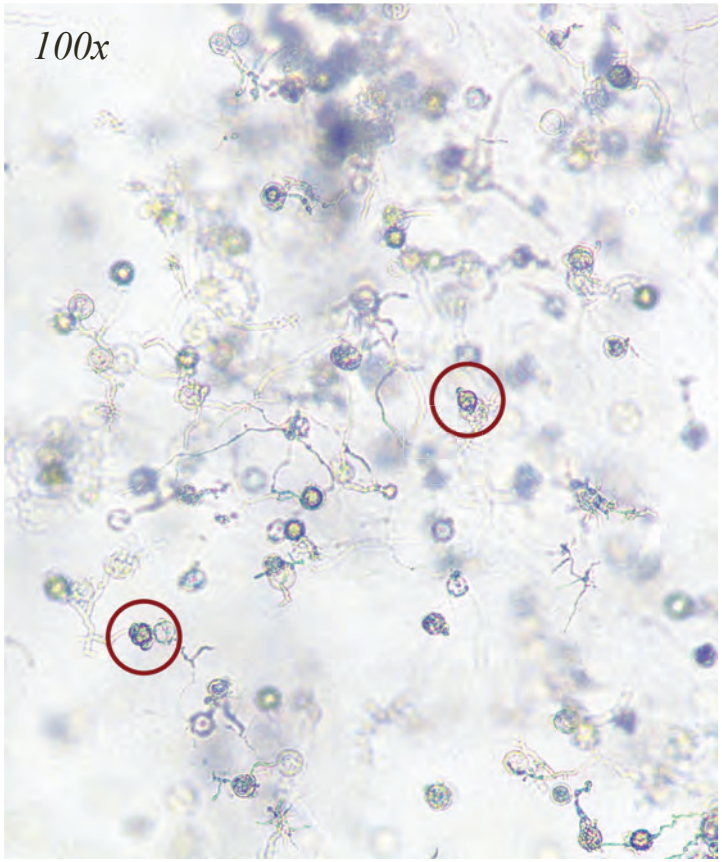
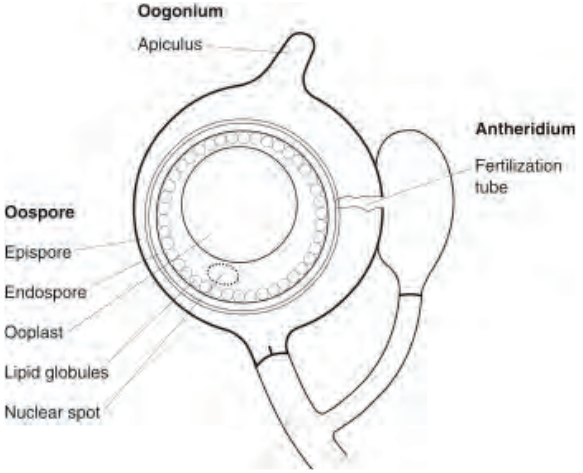
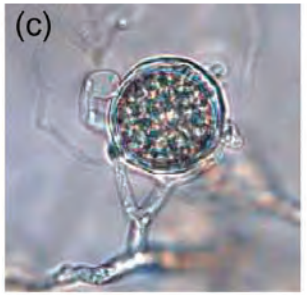


Sandle 2014





Rahman et al. 2015

de Cock et al. 2015

CO
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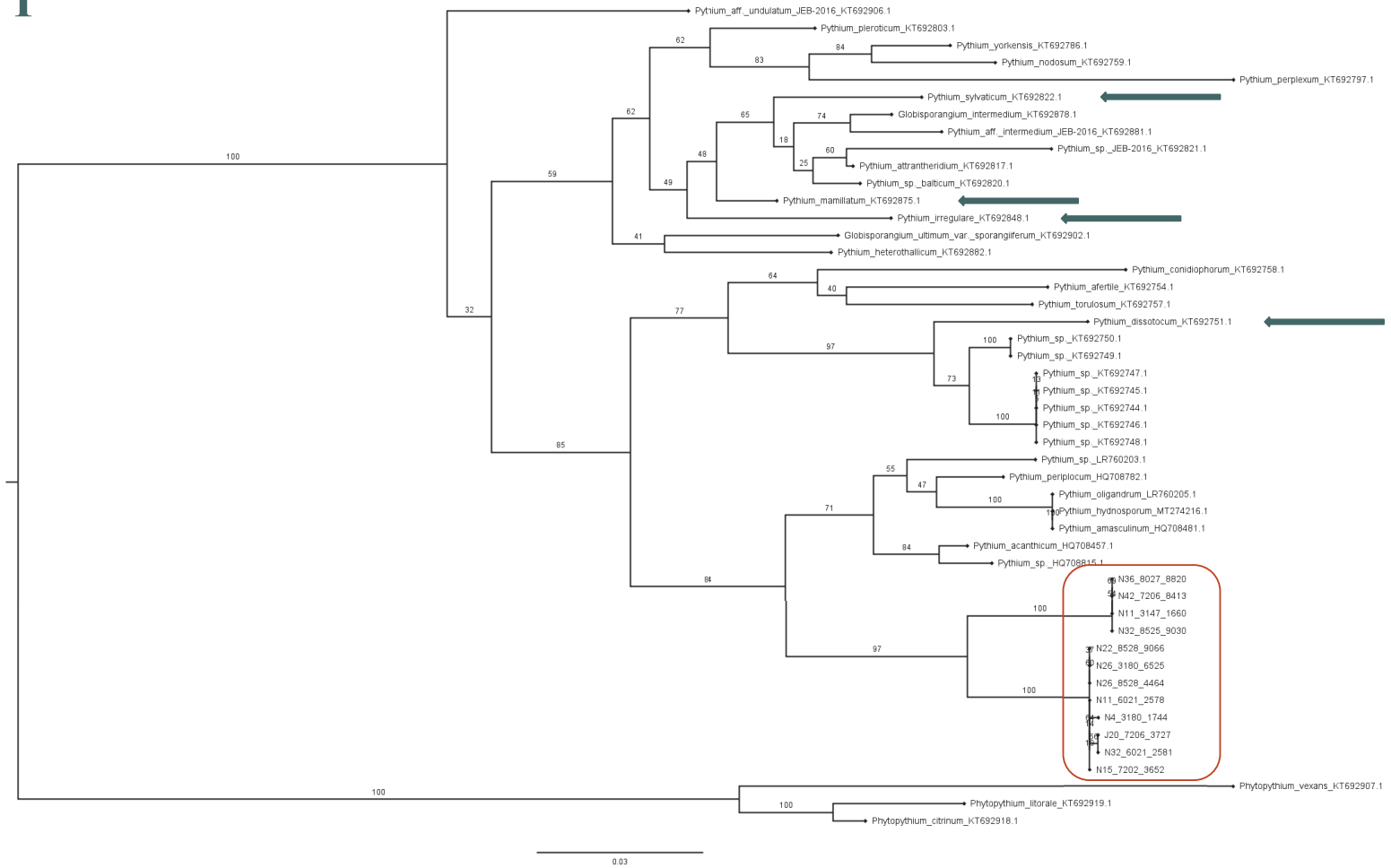
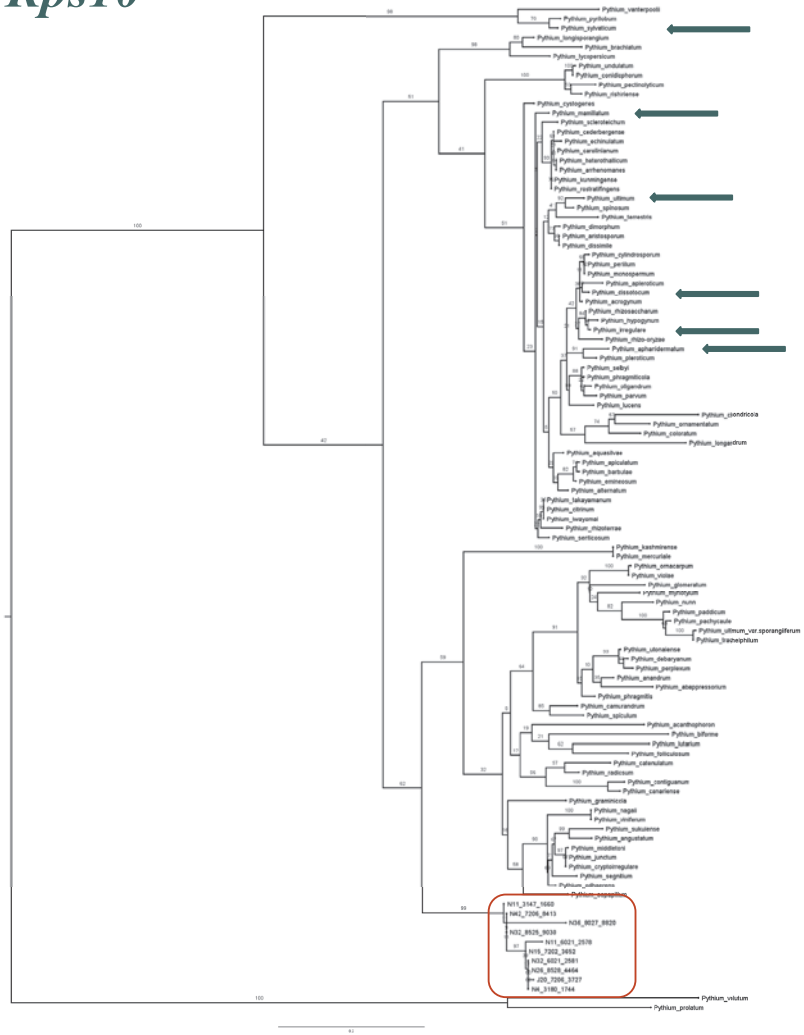

Pythium_aff._undulatum_JEB-2016_KT692906.1
Pythium_pleroticum_KT692803.1
Pythium_yorkensis_KT692786.1
Pythium_nodosum_KT692759.1
Pythium_perplexum_KT692797.1
Pythium_sylvaticum_KT692822.1
Globisporangium_intermedium_KT692878.1
Pythium_aff._intermedium_JEB-2016_KT692881.1
Pythium_sp._JEB-2016_KT692821.1
Pythium_attrantheridium_KT692817.1
Pythium_sp._balticum_KT692820.1
Pythium_mamillatum_KT692875.1
Pythium_irregulare_KT692848.1
Globisporangium_ultimum_var._sporangiiferum_KT692902.1
Pythium_heterothallicum_KT692882.1
Pythium_conidiophorum_KT692758.1
Pythium_afertile_KT692754.1
Pythium_torulosum_KT692757.1
Pythium_dissotocum_KT692751.1
Pythium_sp._KT692750.1
Pythium_sp._KT692749.1
Pythium_sp._KT692747.1
Pythium_sp._KT692745.1
Pythium_sp._KT692744.1
Pythium_sp._KT692746.1
Pythium_sp._KT692748.1
Pythium_sp._LR760203.1
Pythium_periplocum_HQ708782.1
Pythium_oligandrum_LR760205.1
Pythium_hydnosporum_MT274216.1
Pythium_amasculinum_HQ708481.1
Pythium_acanthicum_HQ708457.1
Pythium_sp._HQ708815.1
N36_8027_8820
N42_7206_8413
N11_3147_1660
N32_8525_9030
N22_8528_9066
N26_3180_6525
N26_8528_4464
N11_6021_2578
N4_3180_1744
J20_7206_3727
N32_6021_2581
N15_7202_3652
Phytopythium_vexans_KT692907.1
Phytopythium_litorale_KT692919.1
Phytopythium_citrinum_KT692918.1
0.03

ITS

Rps10



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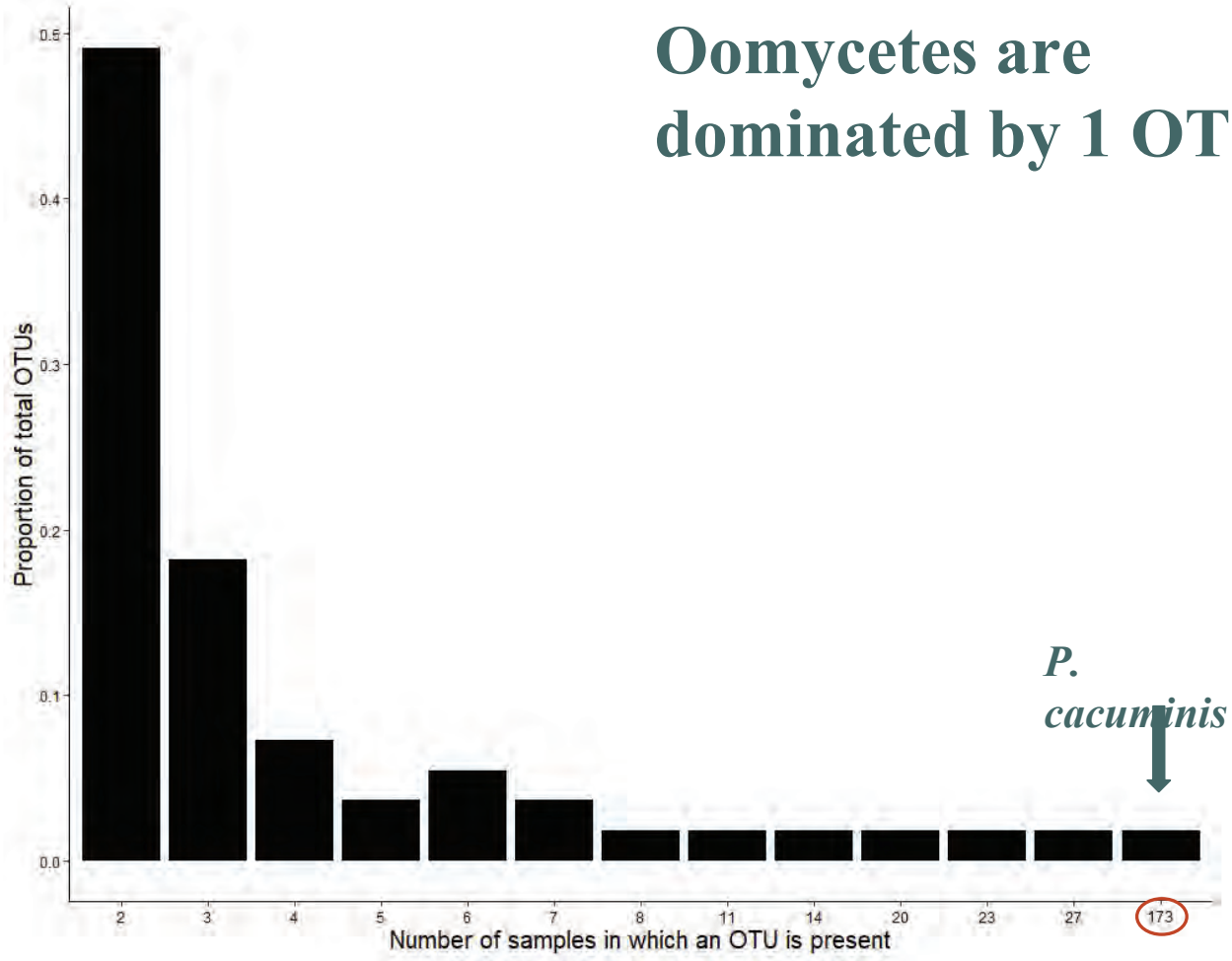
1) Describe community dynamics of foliar fungi and oomycetes of *P. menziesii* on the PNW coast

Oomycetes are dominated by 1 OTU



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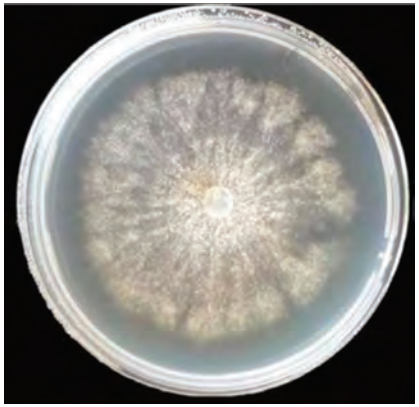
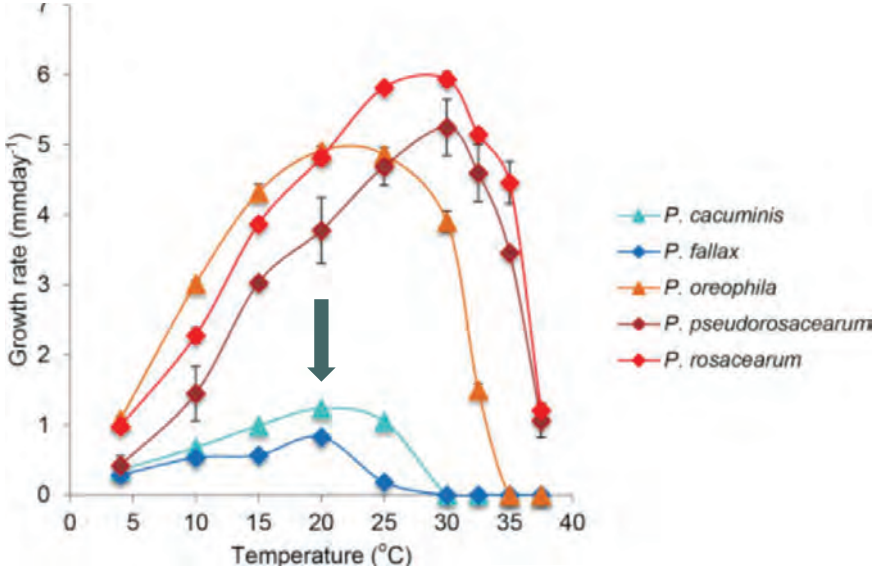
Phytophthora cacuminis



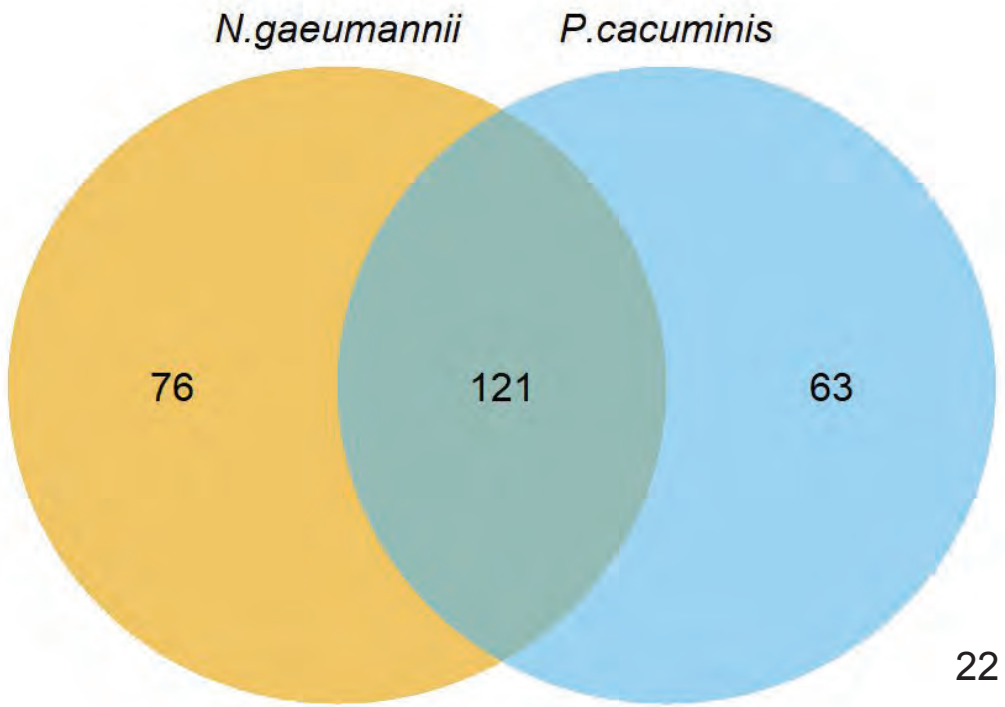
Isolated from asymptomatic *Eucalyptus* root tissue in alpine environments in Australia (Khaliq et al. 2019)

Functional traits suggest adaptation to cold environments
(Redondo et al. 2018)



Figures from Khaliq et al. 2019

4) Investigate any associations between *N. gaeumannii* and oomycetes



4) Investigate any associations between *N. gaeumannii* and oomycetes

Logistic regression (log link)

Response: *N. gaeumannii* presence/absence
Fixed effect: *P. cacuminis* presence/absence
Random effects: Site, Block(Site)

Term	Estimate	Std Error	z value	*p*
Intercept	1.393	0.458	3.047	**0.002**
P. cacuminis	-0.654	0.309	-2.117	**0.03**

***P. cacuminis* present:** $\pi_1 = 0.677$

***P. cacuminis* absent:** $\pi_0 = 0.801$

Thank you

Committee
Jared LeBoldus
Andy Jones
Posy Busby
Dave Shaw

Lab Mates
Patrick Bennett
Kayla Delventhal
Kyle Gervers
Sabrina Heitmann
Devin Leopold
Shawn McMurtrey
Paul Reeser
Kelsey Søndreli
Wendy Sutton
Javier Tabima

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Connie Harrington
Beverly Luke
Chris Poklemba
Brad St. Clair
Dave Thornton

CGRB
Katie Carter
Mark Dasenko
Chris Sullivan

Shrub House
Meredith Jacobson
Kat Lunde
Allie Swartz
Cedar Warman

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Lauren Greer
Livi
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College of Forestry
Swiss Needle Cast Cooperative