2018 Pacific Division Meeting Abstracts

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Sequence diversity of *Impatiens necrotic spot virus*

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Since its discovery, *Impatiens necrotic spot orthotospovirus* (INSV, genus: *Orthotospovirus*, family *Tospoviridae*) has caused significant problems in ornamental and vegetable crops. Reports, however, have shown expanded host ranges of the virus that include field-grown agronomic and horticultural crops. In view of its expanded host range and economic importance, this study was undertaken to examine the genetic diversity of INSV for a better understanding of its epidemiology. For this purpose, we determined the complete sequence of M-RNA and S-RNA genome segments of thirteen INSV isolates from ornamental and vegetable plant species in California, New York and Washington and compared the nucleotide sequences with corresponding sequences available in GenBank. The M-RNA segments varied between 4,950 and 4,961 nucleotides (nt) with a sequence identity between 97.5 and 98.3% compared to previously described isolates. The S-RNA segments ranged from 2,960 to 3,033 bp, with nt identity between 92.2 and 97.5% with those previously characterized. A global phylogenetic analysis of the M- and S-RNA sequences revealed segregation of INSV isolates into two clades with the American isolates distributed in both clades. Further, a comparison of M- and S-RNA segment-based phylograms showed conflicting topology suggesting phylogenetic incongruence due to reassortment between viral genome segments. These results provide an improved knowledge of INSV genetic diversity that has important implications for viral evolution and epidemiology.

Characterizing resistance to metalaxyl resistant *Pythium ultimum* in chickpeas

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Seed and seedling diseases of chickpeas caused by Oomycete *Pythium ultimum* Trow. have been effectively controlled for several decades through the use of fungicide metalaxyl. However, since its first discovery in 2014, seed rot and pre-emergence damping-off of chickpea caused by metalaxyl-resistant *P. ultimum* has become a disease of increasing importance to chickpea production in the U.S. Pacific Northwest. The objective of this study was to (1) develop an efficient screening method (2) identify sources of resistance in chickpea to metalaxyl resistant *P. ultimum* (3) detect QTL associated with disease resistance in chickpea. Genetic markers associated with resistance in chickpea to metalaxyl-resistant *P. ultimum* were detected using an interspecific population consisting of 177 RILs derived from a cross between PI 599072/FLIP 84-92C. Both parents and RILs were screened in growth chamber using soil infested with

oospores of a metalaxyl resistant *P. ultimum*. For identification of QTL, 1032 SNP markers were used to develop a linkage map and association between markers and disease resistance was detected using the composite interval mapping. One highly significant QTL was detected on LG4 with 41.8% of the phenotypic variation. The disease resistant chickpea lines and QTL identified in these studies will be used to accelerate the development of chickpea varieties with improved resistance to seed rot and pre-emergence damping off caused by metalaxyl-resistant *P. ultimum*.

Fungicide resistance occurrence in the blue and gray mold fungi of pome fruit in the Pacific Northwest

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In 2016 and 2017, 320 apple grower lots and 124 pear grower lots were surveyed and we collected 7,000 *Penicillium expansum* and 4000 *Botrytis cinerea* isolates. All isolates were tested for sensitivity to thiabendazole (TBZ), pyrimethanil (PYRI) and fludioxonil (FDL), in addition to pyraclostrobin (PYRA), boscalid (BOSC) and penthiopyrad (PENTH) for *B. cinerea*. Sensitivity tests were conducted using a germ tube length assay on agar media with doses of fungicides known to discriminate resistant from sensitive isolates. Resistance frequencies (RF) in *P. expansum* from apple averaged 25% and 15% for TBZ and PYRI versus 60% and 28% in pear isolates, respectively. Overall, 10 to 13% of *P. expansum* isolates showed reduced sensitivity to FDL but were controlled by the highest label rate. For *B. cinerea*, the highest RF was for PYRI averaging 18% in apple and 35% in pear followed by TBZ with respective RFs of 15% and 26%. For preharvest fungicides PYRI, BOSC, and PENTH, RFs in apple were 30, 22, and 3% versus 26, 14 and 1% in pear, respectively. A majority of *Penicillium* isolates were resistant to one fungicide at time whereas in *B. cinerea*, about 25% were either resistant to only or two fungicides, simultaneously. Resistance is widespread to TBZ and has increased to PYRI in recent years in the PNW whereas reduced sensitivity to FDL is building. Our results show that rotation programs still possible to reduce fungicide resistance development.

Grapevine resistance to Phomopsis foliar and trunk diseases

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Management of grapevine trunk diseases in California is focused on protecting pruning wounds from infection by spores, produced with winter rains. However, recent spore-trapping studies show the causal fungus of the trunk disease Phomopsis dieback, *Diaporthe ampelina*, rarely sporulates at this time. As such, the infection court may not be dormant pruning wounds, but instead green tissues. Indeed, *D. ampelina* causes Phomopsis cane and leaf spot, common on the East Coast, where spores are spread with spring rains to green tissues. A study on hybrid grapes gave us the opportunity to determine if genotypes with resistant green tissues also have resistant woody tissues. Progeny from a cross of European wine-grape *V. vinifera* 'Chardonnay' by North American species *Vitis cinerea* B9 (both resistant to green-tissue infections) segregate for resistance. In the field, 44% of progeny were susceptible, developing severe lesions on stems. A high-density genetic map was used to localize novel qualitative resistance locus *Rda1* from *V. cinerea* B9. In the greenhouse, resistant or susceptible phenotypes were confirmed on green stems of a subset of progeny and both parents. Inoculations to woody stems showed consistent phenotypes in woody tissues. With sibling genotypes that express consistent phenotypes among tissues, we can test hypotheses about infection, for example that the pathogen infects green tissues first, then spreads to woody tissues, where wood cankers form.

Transcriptomic analyses of *Phytophthora parasitica* treated with silver nanoparticles using the RNA-seq approach

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Phytophthora parasitica is a soil-borne pathogen infecting a wide range of plant species in almost 60 plant families, including several economically important crops. Diseases caused by *P. parasitica* can be controlled with fungicides. However, for sustainable management, it is important to discover new effective control methods for controlling *Phytophthora* diseases. Previous studies have indicated various strategies to be useful controlling *P. parasitica*, which include chemical and biological control methods and treatment with nanoparticles. We have previously shown that silver nanoparticles inhibit *P. parasitica* both in vitro and in vivo. To gain molecular insight into how silver nanoparticles work, we conducted transcriptome analysis of *P. parasitica* treated with silver nanoparticles at different times. Computational analyses revealed that the expression levels of thousand genes was modulated by silver nanoparticles. A transient transcriptional upregulation of several genes associated with rRNA synthesis and respiration was observed at 1-hour interval, with significant repression later at 15 hours. Other genes that showed significant differential expression included those involved in amino acid metabolism and cell wall synthesis. In addition, we also observed an overall downregulation of genes associated with general metabolism specially at 1 hr time-points, suggesting that silver nanoparticles inhibit *Phytophthora* growth in a non-specific manner. In conclusion, we propose that AgNPs more likely affect *Phytophthora* by interfering with many vital biological processes, and further research is required to test this hypothesis in vivo.

Protecting a cultural icon and food resource: Current research and status of coconut palm in Guam and the Northern Marianas

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Coconut palm is both an important food resource and a cultural icon for Guam and all tropical islands of the Pacific. Guam in particular among the Insular Pacific islands has been especially hard hit by invasive species due to its centuries-long history as a transportation hub and strategic military location. Health of the overall coconut palm population in Guam is declining more rapidly in recent years since coconut rhinoceros beetle (CRB) was first detected in 2007 in Tumon. Tinangaja, a slow-acting, lethal disease caused by the *Coconut tinangaja viroid*, was previously known only in Guam. Prior to the introduction of CRB, Tinangaja disease was reported to affect 30% of the coconut palm population. Anecdotal reports of the disease in Rota have been confirmed by visual diagnosis by the authors. Results of currently on-going surveys for Tinangaja in Guam and the Northern Mariana Islands will be presented. Additionally, the potential impact of CRB expansion on Tinangaja disease incidence will be presented with an overview of CRB management and biocontrol programs in Guam.

Prevalence of Race 2 Strains of *Verticillium dahliae* causing Verticillium wilt of mint in Oregon

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Verticillium wilt, caused by *Verticillium dahliae*, is the most detrimental disease of commercially grown peppermint (*Mentha piperita*) in Oregon. Two pathogenic races of *V. dahliae* have been identified, but it is not known which race(s) cause(s) Verticillium wilt in peppermint production fields of Oregon. We hypothesized that race 2 *V. dahliae* strains were responsible for Verticillium wilt in peppermint grown in Oregon. Sixty-five isolates of *V. dahliae* were collected from symptomatic peppermint grown in commercial peppermint fields in Oregon during the 2014 and 2015 growing seasons, including 25 isolates from central Oregon, two isolates from the Grande Ronde Valley, and 38 isolates from the Willamette Valley. Identification of *V. dahliae* races was performed using the race-specific PCR primer pairs VdAve1F/VdAve1R (race 1) and VdR2F/VdR2R (race 2). All 65 isolates of *V. dahliae* were classified as race 2 based on PCR using the race-specific primers. An additional 35 isolates of *V. dahliae* collected from mint in various production areas (CA, ID, IN, MI, MT, OR, and WA) were also classified as race 2. These results are consistent with a previous study, which only observed race 2 strains among 16 isolates from mint grown in Washington State. The prevalence of *V. dahliae* race 2 strains observed in association with mint and other crops in the Pacific Northwest implies the importance of developing peppermint cultivars that are resistant to race 2 strains of *V. dahliae*.

Multiple origins of downy mildews and mito-nuclear discordance within the paraphyletic genus *Phytophthora*

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Phylogenetic relationships between thirteen species of downy mildew and 103 species of *Phytophthora* (plant-pathogenic oomycetes) were investigated with two nuclear and four mitochondrial loci, using several likelihood-based approaches. Three *Phytophthora* taxa and all downy mildew taxa were excluded from the previously recognized subgeneric clades of *Phytophthora*, though all were strongly supported within the paraphyletic genus. Downy mildews appear to be polyphyletic, with graminicolous downy mildews (GDM), brassicolous downy mildews (BDM) and downy mildews with colored conidia (DMCC) forming a clade with the previously unplaced *Phytophthora* taxon totara; downy mildews with pyriform haustoria (DMPH) were placed in their own clade with affinities to the obligate biotrophic *P. cyperi*. Results suggest the recognition of four additional clades within *Phytophthora*, but few relationships between clades could be resolved. Trees containing all twenty extant downy mildew genera were produced by adding partial coverage of seventeen additional downy mildew taxa; these trees supported the monophyly of the BDMs, DMCCs and DMPHs but suggested that the GDMs are paraphyletic in respect to the BDMs or polyphyletic. Phylogenetic approaches may be limited in their ability to resolve some of the complex relationships between the “subgeneric” clades of *Phytophthora*, which include twenty downy mildew genera and hundreds of species.

Restoration outplantings of nursery-origin Californian flora are heavily infested with *Phytophthora*

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A survey of 31 restoration sites containing plantings of nursery-origin native Californian flora was conducted between August 2015 and April 2016 in Santa Clara County, California, United States. Samples (191 total) were baited with both pear fruit and rhododendron leaves; a few samples were also baited with *Quercus* leaves or *Salicornia* stems. *Phytophthora* species were isolated from 73 of 191 samples from 23 of 31 sites, yielding 38 species of *Phytophthora*. Up to eight species of *Phytophthora* were baited from single one-liter soil and root samples. *Phytophthora cactorum* was the most common and widespread species, with *P. multivora*, *P. crassamura*, *P. rosacearum* and *P. taxon oaksoil* being frequently encountered. Four novel species were isolated, including *P. taxon mugwort*, a member of a previously uncharacterized subgeneric clade and *P. taxon juncus*, the second species in clade 11. *P. sp. cadmea*, a new provisional species in clade 7 and *P. taxon xguadalupesoil*, a provisional, putatively hybrid taxon in clade 9 were also isolated in the

survey. *Phytophthora quercina*, a species of regulatory concern, was reported for the first time in North America. The survey highlights the potential for widespread *Phytophthora* infestation in outplanted, urban and natural settings, including wildlands.

Wood-rotting Basidiomycetes associated with grapevine trunk diseases in North America

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The trunk disease Esca affects vineyards in all major grape-growing regions of the world. In North America, the causal fungi that are best studied are members of division Ascomycota *Phaeoconiella chlamydospora* and *Phaeoacremonium* species. Fungi in division Basidiomycota (species of *Auricularia*, *Fomitiporia*, *Inonotus*, *Phellinus*, *Stereum*, and *Trametes*), which are thought to be secondary colonizers, are reported from Australia, Europe, New Zealand, South Africa, and South America. However, there are no studies testing this sequence of colonization. With only two reports of such Basidiomycetes in North America, we had to, first, develop a collection to identify the causal fungi and, second, carry out controlled inoculations to test their pathogenicity. A survey of 22 vineyards with the characteristic white-rot symptom in Texas and California revealed five species: *Coprinellus radians*, *F. langloisii*, *F. polymorpha*, and *Inonotus andersonii*. A majority of wood samples yielding these isolates (160 of 174) were collected from vines showing the diagnostic leaf symptoms of Esca. *Fomitiporia spp.* are different from that reported in Europe, *F. mediterranea*. Additional taxa within *Tropicoporus* and *Fomitiporia* represent undescribed species. In the greenhouse, wood discoloration was present among plants inoculated with all isolates. Leaf symptoms were less common, but varied from a marginal or interveinal scorching to the diagnostic leaf symptom of Esca.

Selective media for recovery of *Graphium euwallaceae* and *Paracremonium pembeum* from polyphagous shot hole borer field samples

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Fusarium Dieback (FD) is a vascular disease that causes branch dieback and tree mortality on a wide variety of woody hosts in CA. The disease results from inoculation of fungal mutualists (*Fusarium euwallaceae*, *Graphium euwallaceae*, and *Paracremonium pembeum*) vectored by an invasive ambrosia beetle (*Euwallacea sp. nr. fornicatus*) named Polyphagous Shot Hole Borer (PSHB). The vector relies on its fungi, mainly *F. euwallaceae*, to support its life cycle inside their galleries in woody hosts, but when isolating from field samples, *G. euwallaceae* and *P. pembeum* are often difficult to recover due to the dominance of *F. euwallaceae* in its vector and/or secondary fungal contamination in infested wood. In this study, various fungicides were tested *in vitro* to determine effective fungicide concentrations for developing selective media. Potato dextrose agar plates amended with the fungicides azoxystrobin, flutriafol, tebuconazole, and thiabendazole were tested at various concentrations (0.01, 0.1, 1, 10, 50, and 100 ppm) against PSHB mutualists to determine minimum effective concentrations for mycelial growth and spore germination. Thiabendazole inhibited both mycelium and spores of all fungal mutualists at a 100 ppm concentration, while tebuconazole at 80 ppm selected for *G. euwallaceae*, and thiabendazole at 10 ppm selected for *P. pembeum*. The two selective media were shown to select for these fungi from infested wood specimens as well as spore suspensions made from PSHB specimens.

Fusarium circinatum infects grasses and pines at Pt. Reyes National Seashore

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Fusarium circinatum is a soil borne fungus and causal agent of pitch canker disease in pines and Douglas fir. The fungus can also infect grasses without causing disease, meaning grasses are hosts for *F. circinatum* without conspicuous symptoms. A survey of grasses growing in pitch canker infested pine stands at Pt. Reyes National Seashore recovered *F. circinatum* from *Pinus muricata* and the grass species *Bromus carinatus* and *Holcus lanatus*. The fungal isolates from grasses share vegetative compatibility with those of pine, suggesting a shared lineage between grass and pine isolates. We inoculated greenhouse-grown *Pinus radiata* with our field isolates, and isolates from all three host species manifested symptoms of pitch canker disease. We undertook experiments inoculating the California native grass *B. carinatus* with *F. circinatum*. Preliminary data suggest *F. circinatum* can sporulate on senescing *B. carinatus* leaves in a moist chamber. We are continuing inoculation experiments of *B. carinatus* to examine if *F. circinatum* can propagate on senescing grass tissues, raising the possibility that the fungus can jump from a grass host to a pine host. Given the lack of symptoms associated with grass infections, grasses may represent an additional inconspicuous source of inoculum for susceptible pines.

Population genetic structure of *Claviceps purpurea* in cool-season grass seed crops of Oregon

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Ergot, caused by *Claviceps purpurea*, is a primary disease concern in irrigated cool-season grass seed production systems of the U.S. Pacific Northwest. However, the understanding of the genetic diversity and population structure of *C. purpurea* is limited. Two hundred-twenty six isolates were obtained using a hierarchical sampling strategy from two fields each of Kentucky bluegrass (Madras OR, $n = 102$) and perennial ryegrass (Hermiston, OR, $n = 124$) and characterized using 12 microsatellite markers. A total of 194 unique multilocus genotypes were identified and the majority of the clones were identical to their peers collected from the same seed head, indicating a role of conidia (honeydew) in secondary infections within seed heads. There were moderate levels of genotypic diversity (*H*

= 3.43 to 4.23) and gene diversity (H_{exp} = 0.45 to 0.57) within fields. After clone correction, analysis of molecular variance revealed that 66% of the genetic variation was between the two *C. purpurea* isolates collected from the same seed head of individual plants. Genetic differentiation was also observed among populations from different hosts (21%), but was confounded by geography. The standardized index of association ranged from 0.005 to 0.122 among the four populations, suggesting differences in the relative importance of ascospores and conidia among the four fields. These results provide insights into the epidemiology of ergot in cool-season grass seed crops of Oregon.

Novel sequencing reveals diagnostic assay of MAT1-1 and MAT1-2 mating type idiomorphs in *Botrytis paeoniae*

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Botrytis paeoniae causes gray mold infection of herbaceous and tree peony (*Paeonia* spp.), and is the foremost challenge faced by the burgeoning peony industry in the northwestern United States. No evidence for sexual reproduction has been documented for this fungus. As part of a larger effort to determine genetic diversity of the northwest United States *B. paeoniae* population, we sought more detail of its mating type (MAT) loci. A draft whole-genome sequence of *B. paeoniae* contained a region homologous to MAT1-1 in *Botrytis* species *B. cinerea* and *B. elliptica*, and led to the successful sequence of the 4.1 kb region in several *B. paeoniae* isolates. Guided by sequence alignments of published MAT1-2 idiomorphs of other *Botrytis* species, primers were developed to amplify the MAT1-2 locus in *B. paeoniae*, and the previously uncharacterized 4.2kb sequence was determined by primer walking. In total, four MAT1-1 isolates and five MAT1-2 isolates showed 100% similarity within the two mating types and 64% similarity between them. A 157bp sequence gap present near the 3-prime end of MAT1-2 and not MAT1-1 allowed for the design of a robust diagnostic PCR and agarose gel assay to determine mating type. Our work represents the first time both mating type loci have been characterized in *B. paeoniae* and indicates that they exist in sympatry. Further, this work represents the third *Botrytis* species for which two mating idiomorphs have been characterized.

Modeling the spatial distribution of the potato cyst nematode *Globodera pallida* in Idaho

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The potato cyst nematode *Globodera pallida* is an economically important potato pest and is globally regulated. *Globodera pallida* is an obligate sedentary endoparasite that requires a living potato plant to complete its life cycle. This nematode originated from the Andean region of South America and was introduced into Europe around 1850 before spreading throughout the world. In highly infested fields, *G. pallida* can cause up to 80% yield loss. *Globodera pallida* was first found in the U.S. in 2006 at a potato processing facility in Idaho. A spatial analysis was used to understand the distribution of *G. pallida* in Idaho and to evaluate the potential threat of *G. pallida* for entry into new areas. Spatial point pattern analysis indicated that *G. pallida* infested fields are spatially aggregated as an ellipsoidal-shaped cluster. Prior to the establishment of containment measures, the spread of *G. pallida* followed a contagion effect scenario where infested fields contributed to the infestations of nearby fields, probably by agricultural machinery or plant materials contaminated with soil infested with *G. pallida*. Spatial interpolation indicated that the containment measures disrupted the spread momentum of *G. pallida* as illustrated by the presence of weak spatial autocorrelation in semivariogram analysis. The current *G. pallida* infested areas were estimated to 1,233 ha in 2017 and were located in southern Idaho. We determined that our conceptual framework and spatial analysis support the eradication of *G. pallida* from Idaho. The tools and techniques used in this study should contribute to the development of spatial model analysis for other invasive plant-parasitic nematodes.

Survey for *Ceratocystis fimbriata* on *Syngonium* species in Hawaii

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Ohia (*Metrosideros polymorpha* Gaud.) is a foundational tree of the Hawaiian rainforest ecosystems that has been rapidly dying, due to a disease caused by, *Ceratocystis fimbriata*. Currently, this disease has only been found on the Big Island of Hawaii. *Ceratocystis fimbriata* was earlier reported on *Syngonium* from Hawaii. It is vital to determine if this pathogen is on other islands on diseased *Syngonium* and to determine its host range. A survey of the island of Oahu and Big Island was conducted. Twenty-two different locations on Oahu were surveyed. Washed samples were dissected into smaller pieces, surface disinfected with 0.6% NaOCl and plated on water agar or incubated under moist conditions. Single hyphal tips of the mycelium growing out from the infected samples or ascospore masses protruding out of perithecia were transferred to 10% vegetable juice agar. *Ceratocystis fimbriata* was not isolated from any samples collected on Oahu. Out of three nurseries surveyed on the Big Island, *C. fimbriata* was isolated from samples collected from two nurseries. Symptoms were black rotting of the nodes and roots and dark lesion on petiole. Pathogenicity of *Ceratocystis fimbriata*, isolate 3474, was tested on *Syngonium* and ohia. Spore suspensions at 1×10^5 endoconidia per ml sterile distilled water were sprayed on unwounded *Syngonium* plants, whereas ohia were wounded with a sterile scalpel and sprayed. Disease symptoms were observed on inoculated *Syngonium* 3 days after inoculation (DPI) and plants were severely wilted after 11 DPI. *C. fimbriata* was reisolated from diseased *Syngonium*. No symptoms have been observed on ohia to date and ohia plants are still under observation.

Wheat variety-specific recruitment of bacterial communities towards *Rhizoctonia solani* AG8 suppression

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The root rot disease in wheat caused by *Rhizoctonia solani* AG8 has remained challenging to control due to lack of resistance in the wheat germplasm and the constraints of using biological control agents. With this, identification of specific wheat varieties that recruit bacterial communities involved in soil-borne disease suppression would provide a more sustainable approach than other management practices. A pilot study by our group showed differences in rhizosphere microbiome among wheat varieties grown in the field for two successive seasons. To accelerate analyses of microbiome differences among wheat varieties, six winter wheat varieties were planted for three 35-day cycles in the growth chamber to mimic three seasons of wheat cropping in Pullman and Lind soils. Amplicon sequencing of 16S rDNA was done at the end of the third cycle. The remaining soils cultivated with the six wheat varieties were then inoculated with *Rhizoctonia solani* AG8-C1. Alpowa seeds were planted and root disease was scored after 14 days to determine “suppressiveness”. The 35-day cycles have resulted to 85 and 53 bacterial taxa that are differentially abundant among the six wheat varieties in Pullman and Lind soil, respectively. However, no significant differences found in the root disease scores in soils cultivated with the six wheat varieties. Further experiments are needed to determine whether three 35-day cycles are enough to recruit putative disease “suppressive” microbiomes.

***Botrytis cinerea* on faba bean in Washington State**

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Beginning in 2013, plants of *Vicia faba* grown near Mt. Vernon, WA bore moderate to severe symptoms of chocolate spot. Isolates P11V12, P31VO9, P34V12, and Vfaba2-1-ht1 were recovered from symptomatic plants in 2015. DNA sequences from partial RPB2 and G3PDH genes placed the isolates in a clade with *Botrytis cinerea*, *B. fabae*, *B. pelargonii* and *B. eucalypti*. The former two species are widely documented as agents of chocolate spot, whereas *B. eucalypti* is documented only on *Eucalyptus* spp. in China; and *B. pelargonii*, documented from *Pelargonium* spp., may be conspecific with *B. cinerea*. On the basis of colony characters, conidial size, host and membership in the above clade, the isolates are provisionally assigned to *B. cinerea*. Greenhouse-grown plants of *V. faba*, accession PI291010, were independently challenged with conidial inoculum of each isolate in repeated trials, resulting in symptoms of chocolate spot and re-isolation of the fungus in each trial; controls were symptomless. *Botrytis* isolate PS036RB from a symptomatic seedpod grown near Pullman, WA in 2014, in the same clade as above and also assigned to *B. cinerea*, has been pathogenic in a preliminary trial. We believe this is the first confirmation of chocolate spot on faba bean in Washington State. The presence in Washington State of the other agent of chocolate spot, *B. fabae*, remains conjectural. Faba bean is an emerging specialty crop in Washington State.

Factors that influence the efficacy of steam mitigation for *Phytophthora ramorum* in soil

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Steaming has become the Confirmed Nursery Protocol mitigation treatment of choice for soil at *P. ramorum* positive (Pr+) sites in Washington State. Eradicating *P. ramorum* from soil requires a temperature of 50°C for at least 30 minutes at 30 cm depth. WSU has provided technical support to APHIS and WSDA using a Sioux SF-20 steamer (steam output 680 lb/hr) to do soil steaming at Pr+ sites. Replicated trials of steaming soil under varying temperature and moisture conditions were conducted at WSU-Puyallup. Heating and cooling times to 50°C were evaluated on sandy loam, silt loam, and nursery gravel bed soils under conditions of saturated (20–50%, vol/vol), field capacity (10–35%), and residual water content (1–10%). Stepwise regression of soil heating data indicated that starting soil temperature was not as important as soil type, depth, and moisture content. Higher ambient temperatures prolonged the cooling phase and the time the soil is above 50°C, especially at lower depths. The 50°C threshold while steaming heavy soils during the cool wet season is difficult to achieve and such effort is unlikely to be cost-effective. Well-drained soils with higher sand content could be steamed efficiently throughout the year. Eradication of *P. ramorum* from a site reduces the risk of the inadvertent movement of this pathogen and may prevent a quarantine being imposed that would affect trade outside of the region.

Using chickpea polygalacturonase-inhibiting protein as a tool to elucidate molecular mechanisms of chickpea interactions with various pathogens

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Chickpea, an important rotational crop in the US Pacific Northwest, encounters a number of diseases caused by species of *Fusarium*, *Macrophomina*, *Pythium*, *Rhizoctonia* and *Sclerotinia*. At some point of the infection process these pathogens produce polygalacturonases (PGs) in degrading plant cell walls. To defend themselves, plants deploy polygalacturonase-inhibiting proteins (PGIPs) that inhibit fungal PGs. More than 170 complete or partial PGIPs have been identified in various plants based on sequence identity, but only a few of them have been empirically shown to encode proteins with PG-inhibition activities. In order to explore the usefulness of chickpea PGIPs in studying diseases resistance mechanisms, we have cloned and sequenced the complete open reading frame of chickpea PGIP (*CaPGIP1*) from cultivar ‘Dwelle’, which had one synonymous substitution from the known chickpea genome sequence and was found to have conserved domains like leucine-rich repeats and ribonuclease inhibitor (RI)-like subfamily. Phylogenetic analysis suggested a close relationship of chickpea PGIP with those of other legume species. This cloned *CaPGIP1* is being expressed in yeast cells and purified to confirm its functions in inhibiting and interacting with PGs of chickpea pathogens and also to be expressed in *Arabidopsis* to study its functions in disease resistance.

Disease gradients of late blight of potato from infrared images of infected fields

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Epidemics caused by aerially dispersed pathogens include some of the most widespread and economically important diseases, such as late blight of potato caused by *Phytophthora infestans*. These epidemics have been shown to follow similar disease gradients, with high local saturation and non-exponentially bound tails. Understanding dispersal and disease gradients can improve efficacy and timing of control measures. Here, infrared images of late blight-infected fields from the Columbia Basin in Washington were examined and converted to black and white, from which pixel discoloration counts were used as a proxy for mean incidence per row. From these images, disease gradients from the focus to the furthest row with a positive number of discolored pixels were cropped, then fit by the modified inverse-power distribution (MIPD), $y = a(x+c)^{-b}$, which has been widely used to describe aerial dispersal. The MIPD best-fit of $y = 12.22(x+140)^{-5.63}$ ($p < 2.2 \times 10^{-16}$) has a considerably steeper b and a larger c than previous reports of aerial dispersal, perhaps due to failing to capture the tail of the dispersal gradient. Therefore, multiple non-linear distributions were fit to the data, which can handle 0s, so a larger portion of the tail could be included. The gamma distribution provided the best fit: $y = (0.71^{0.11} x^{0.89} e^{-0.71x}) / \Gamma(0.11)$. This study demonstrates the importance of assessing the tails of dispersal gradients and its results can improve spatially explicit forecasting models.

The IPM concept of thresholds in plant disease management

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The principles of integrated pest management (IPM) originated from studies of aphids in alfalfa in the 1950s. Core concepts include accurate pest identification, understanding pest biology to design prevention and avoidance tactics, monitoring for pests, and using of efficacious suppression methods when needed. In insect management, suppression tactics are employed when populations exceed a threshold which is tracked by ongoing monitoring. Thus, some IPM practitioners consider thresholds a prerequisite for well-designed IPM programs. This view is parochial since thresholds do not work as well for plant pathogens and weeds as they do for arthropods. Although, thresholds have been successfully used for single cycle, soilborne diseases, they are less appropriate for insect-vectored and polycyclic, foliar diseases. The lack of thresholds for these diseases results from differences in pest biology and not from a lack of IPM understanding in plant pathology. More broadly, thresholds are just one method of evaluating monitoring data to determine timing of suppression tactics. Observation of environmental conditions, employing weather-based forecasting models, and scouting for insects that vector plant diseases are all examples of monitoring activities that determine when to employ suppression tactics and integral to successful IPM in plant pathology.

The stem and bulb nematode, *Ditylenchus dipsaci*: An enigmatic species of increasing importance in North America

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The stem and bulb nematode, *Ditylenchus dipsaci* (Kuhn) Filipjev, is the most widespread and economically important of the relatively few species of plant parasitic nematodes that cause diseases of the above-ground portions of plants. Approximately 500 wild and cultivated plant species are recorded hosts for *D. dipsaci*, which is thought to be comprised of at least 30 different races defined on the basis of host ranges. In western North America, *D. dipsaci* is best known as a significant pest of alfalfa, garlic, onion and ornamental bulbs. Despite being very widespread in temperate regions, the nematode is nonetheless a significant regulatory concern and is listed as a quarantine pest by many countries. This presentation will review the status of the nematode in North America, including recent outbreaks on garlic in Midwestern and eastern parts of the US and Canada, as well as unique aspects of the biology of the nematode such as its ability to undergo anhydrobiosis. The presentation will also explore emerging gaps in research on *D. dipsaci*, including gaps in (1) understanding of *Ditylenchus* taxonomy and the development of diagnostic protocols needed to deal with regulatory issues; and (2) understanding of the influences of environmental conditions, climate change and changes in crop management practices on the epidemiology of *D. dipsaci*.

Grapevine trunk pathogens in California associated with the apoplectic stage of Esca

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Summer 2017 was notable for numerous reports statewide of the Esca symptom known as ‘apoplexy’, when entire grapevines suddenly wilt and die from this trunk disease. These reports revealed gaps in the research on Esca relative to the dieback-type trunk diseases, Botryosphaeria-, Eutypa-, and Phomopsis diebacks. Filling these gaps is important for making accurate recommendations to growers about managing all trunk diseases, including Esca. The Esca pathogens, like other trunk pathogens, reside in the wood. Esca symptoms of leaf interveinal necrosis and fruit spots typically occur on shoots growing from the same cordon or cane each year, with variable interannual severity. Some years, though, instead of individual shoots showing symptoms, the entire vine dies from apoplexy. What triggers this change from isolated symptoms one year to total vine collapse the next year is not known, but consistent factors are wet spring weather and high temperatures before veraison, both of which happened in 2017. From our survey of symptomatic wine grapes, the majority on Freedom rootstock, we isolated *Phaeoconiella chlamydospora* and *Phaeoacremonium* spp., which have been shown as causal agents of Esca and are reported from northern California vineyards. The most virulent of these fungi, *Phaeoacremonium minimum* and *Phaeoconiella chlamydospora*, made up less than 1% and 30% of samples, respectively, suggesting other *Phaeoacremonium* species may be more important than previously thought.

Grapevine trunk pathogen *Neofusicoccum parvum* tolerates host phytoalexin phenolic compounds

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Trunk pathogens are fungi that infect grapevine wood primarily through pruning wounds and establish localized infections (wood cankers), which are often lethal to adjacent fruiting positions. *Neofusicoccum parvum* and *Diplodia seriata* (causal fungi of Botryosphaeria dieback) and *Eutypa lata* (causal fungus of Eutypa dieback) all cause cankers, but vary in virulence. We examined the potential role of anti-microbial phenolic compounds, which have been shown to increase in response to wounding and infection, in the ability of each fungus to colonize the host. In vitro assays were used to test the effects on fungal growth and fungal alteration of the following phenolic compounds: gallic acid, epicatechin, rutin, and piceid. *N. parvum* was either uninhibited or promoted by the compounds, whereas biomass of *D. seriata* and *E. lata* was up to 50% less than that of cultures grown in unamended media. *N. parvum* more efficiently utilized rutin and gallic acid, which were eliminated to nearly non-detectible levels within 2 or 5 days, respectively, compared to 15 and 21 days for *D. seriata* and *E. lata*, respectively. *N. parvum* eliminated piceid within 2 days, whereas *D. seriata* and *E. lata* accumulated resveratrol at 2 and 7 days, respectively, before degrading it. Epicatechin was utilized efficiently by all three fungi. The ability of *N. parvum* to tolerate these phenolics or utilize them as carbon sources may contribute to its greater virulence compared to *D. seriata* and *E. lata*.

Validation of *Xylella fastidiosa* synthesized gene fragments for use as a qPCR control with inserted BamHI cut sites for downstream quality control

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In diagnostic work of plant pathogens, obtaining timely positive controls can be challenging due to USDA permit constraints to move live organisms, upkeep and culturing, or locating sources which have the material readily available. Our lab has validated the use of a specially designed synthetic gene fragment for qPCR detection of the 16S rDNA gene of *Xylella fastidiosa*. In addition to detection, our synthesized control was designed with a BamHI restriction enzyme site not present in the target sequence, enabling us to clearly distinguish a true positive result from that of our synthetic control using digestion and gel electrophoresis. The inserted restriction site was selected based upon a conserved area found within the 16S rDNA alignment of all known strains of *X. fastidiosa*. Our work shows that synthesized gene fragments are consistent, cost effective, easily manipulated, and can be designed to contain built in quality control measures. These fragments can be designed to contain multiple targets from different assays, including internal control sequences for multiplexed reactions. Another benefit to this cost effective method is having abundant quantities of template to conduct optimization, validation, and diagnostic testing for the targeted pathogen. We have successfully implemented the use of such synthetic controls for additional pathogens such as: *Phytophthora kernoviae*, *Phytophthora ramorum*, *Candidatus Phytoplasma* spp., and *Cylindrocladium buxicola*.

Host plant resistance to *Macrophomina* crown rot and *Verticillium* wilt in strawberry cultivars and elite selections

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The soilborne pathogens *Verticillium dahliae* and *Macrophomina phaseolina*, causal agents of Verticillium wilt and Macrophomina crown rot, have emerged as prevalent pathogens in California strawberry production. Both pathogens produce microsclerotia as survival structures, making these diseases difficult to manage without soil fumigation. Host resistance will be a key factor for future disease management. Germplasm from five private and public breeding programs were evaluated for disease resistance in field trials consisting of 90 cultivars and elite selections in replicated 20-plant plots. In the case of *Verticillium*, the trial was conducted in a field naturally infested with *V. dahliae* (25 CFU per gram of soil). For *Macrophomina*, plants were inoculated post-transplant using a cornmeal-sand inoculum at 5 g/plant. Resistance was measured by recording plant mortality over two production seasons (2016–17 and 2017–18). Data from the 2016–17 trial suggests a wide range of resistance to each pathogen among germplasm in all breeding programs; cultivars showing evidence of exceptional resistance within the *Macrophomina* trial were Osceola, Manresa, Megan, Grenada and Laredo, while those most resistant to *Verticillium* were Camino real, Marquis, Petaluma, San Andreas and Albion. The goal of this research is to identify cultivars that can be successfully grown in fields with a history of soilborne disease and to guide breeding programs in future development of host resistance.

Activities of *Fusarium circinatum* in soil

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Fusarium circinatum is best known as the cause of pitch canker, a disease that affects many pine species and Douglas-fir. Typical symptoms of pitch canker include canopy dieback and resinous lesions on the main stem of the tree. *Fusarium circinatum* can be seedborne, which provides a mechanism for long-distance dispersal. Sowing infested seed allows the pathogen to become established in soil. Although *F. circinatum* does not produce chlamydospores, it can survive for at least 12 months in either wet or dry soil. Spores of the pathogen can germinate and infect roots of susceptible pines, without causing visible damage. Seedlings will become symptomatic

only after growth in the root system reaches the soil line, where the pathogen can girdle the stem, killing the seedling. However, the pathogen may remain alive in the root system for at least 12 months without a visible effect on the plant, either above or below-ground. Consequently, infected seedlings may be distributed for use in plantations or as landscape trees. Examining roots for evidence of infection is not a reliable method of detecting an infected seedling, because root death occurs only after decline of the shoot. Cryptically infected seedlings may fail in out-plantings, while introducing the pathogen to new locations. Although root infections by *F. circinatum* may be seen as latent, with the potential for future damage, mild infections can actually enhance growth and induce resistance to stem infections.

Pre-harvest management of Phytophthora brown rot to prevent trade restrictions of California citrus

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Brown rot, caused by *Phytophthora citrophthora* (Pc), *P. syringae* (Ps), *P. parasitica*, and *P. hibernalis*, is an important disease of citrus in California. *P. syringae* and *P. hibernalis* are quarantine pathogens, and their detection resulted in closures of some California export markets. Asymptomatic fruit at the time of packing may develop the disease during storage and transit, only to be detected during port-of-arrival inspection. For many years, only copper and phosphonate fungicides were available for pre-harvest foliar application for managing brown rot. Field trials were conducted with registered and new (mandipropamid - MAN, oxathiapiprolin - OXA) fungicides from 2014 to 2018. Following one or two applications per harvest season, fruit were sampled periodically over two months, inoculated with Ps or Pc, and incubated. MAN and OXA were highly effective and significantly reduced disease incidence from the control over each sampling period with different total amounts of rainfall. Copper was mostly highly effective, whereas the efficacy of PP declined during the first five weeks or following major rain events. MAN and OXA premixtures and their rotation with copper applications were also highly effective and will provide critical resistance management strategies. The new fungicides will reduce over-usage of copper and minimize phytotoxicity and soil and water contamination. OXA was federally registered in 2018, and registration of MAN is pending through IR-4.

Practical management for Soilborne wheat mosaic virus: An emerging disease in the dryland Pacific Northwest

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Soilborne wheat mosaic virus (SBWMV) was discovered in the dryland wheat production zone of the PNW in 2008. Since SBWMV is an emerging disease in the region, yield loss and distribution is unknown. Limited genetically resistant winter wheat cultivars are available, as breeders have recently begun to incorporate major resistance into PNW-adapted cultivars. In response to this emerging disease, we documented current SBWMV distribution and conducted yield loss estimates in dryland conditions. In addition, potential management techniques, such as nitrogen applications and variety blends were tested. Preliminary data suggest SBWMV is a yield-limiting disease that is spreading in the dryland PNW. Significant yield loss of 37.3 bu/A ($p < 0.001$) was documented in commercial winter wheat fields in the 2017 season. Since the first discovery in 2008, an expansion of the affected area from a 3-mi radius, to an over 25-mi radius was documented. Preliminary results indicate an additional nitrogen application is not effective to prevent yield loss due to SBWMV. Preliminary results also indicate resistant/susceptible variety blends do not significantly differ in yield compared to pure stand resistant varieties under severe SBWMV conditions. Therefore, variety blends may be an excellent practical management solution for SBWMV while resistant cultivars are limited. This work is part of an ongoing effort to provide practical management solutions to an emerging soilborne virus.

Does it take two to tango?: Interaction of *Fusarium pseudograminearum* and *Pratylenchus neglectus* on wheat

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Pathogens are often studied in an isolated manner to gain an understanding of a single pathogen's effect on a crop. However, in a practical field setting, pathogens rarely exist in isolation. *Fusarium pseudograminearum* and *Pratylenchus neglectus* are two yield-limiting pathogens with overlapping distribution in the 2-billion-dollar dryland wheat production system of the PNW. The goal of this greenhouse study was to understand how *F. pseudograminearum*, the causal agent of Fusarium crown rot and the root lesion nematode, *P. neglectus*, interact and influence disease symptomology when co-inoculated on wheat. Winter and spring wheat varieties were inoculated with ~400 mixed stage *P. neglectus* at 10 days after planting. At 12 days after planting, 150 mg of millet seed infested with *F. pseudograminearum* was distributed on wheat seedling crown tissue. Thirteen weeks after planting, plants were destructively harvested and rated for plant height, dry plant weight, Fusarium crown rot damage, and dry root weight. *P. neglectus* reproduction was quantified by extracting nematodes from roots and soil. Results indicate lower *P. neglectus* reproduction when co-inoculated with *F. pseudograminearum*, compared to treatments inoculated with *P. neglectus* alone ($p < 0.001$). This result may be due to reduced root mass in *F. pseudograminearum* inoculated treatments compared to uninoculated controls ($p = 0.0005$). Results indicate no significant difference in Fusarium crown rot severity when co-inoculated with *P. neglectus* compared to treatments inoculated with *F. pseudograminearum* alone ($p = 0.69$). This study is part of an ongoing effort to evaluate risk factors and practical disease management for dryland wheat producers.

LOOK UP FOR PHYTOPHTHORA! Phytophthora as a foliage, shoot, and stem pathogen of forest trees

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Phytophthora is often pigeon-holed as a soil and water-borne pathogen destructive on agronomic crops. That was certainly my mindset until Sudden Oak Death forced my attention to the tree tops. Water is indeed essential to the disease cycle, but zoospores swim in a water film on leaf surfaces as well as in soil, witness *P. infestans*, and why not 50 meters above ground? Foliar *Phytophthoras* are diverse, locally numerous, and apparently indigenous in many forests of the world. On four continents some species are also invasive, causing destructive forest diseases that threaten whole ecosystems. Examples of foliar *Phytophthoras* can be found in most of the phylogenetic clades of the genus, usually accompanied by the morphological adaptation of deciduous sporangia. *Phytophthora* Clade 3 is seemingly an exclusively foliar group, perhaps indigenous to western North America, with soil populations arguably representing the hazards of falling from high places and epidemiologically insignificant. *Phytophthora lateralis*, and *P. ramorum*, of Clade 8, illustrate the invasive potential of this adaptation, and the challenges that aerial spread poses to disease management. Both species are subject of intensive disease management efforts that have been surprisingly successful, despite the epidemiological challenges posed by their life histories.

Adapting soil moisture models to improve epidemiological modeling of soilborne plant disease

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The epidemiology of soilborne plant pathogens is a realm of study often underutilized when seeking to improve sustainability in agriculture. The reasons for this are likely varied. However, as growers seek more means of improving sustainability in food and fiber production in an ever-changing climate, predictive epidemiological models can provide insight in how to maintain high yield in an enduring manner. In an ongoing study, we seek to add to the toolbox of integrated pest management by more definitively characterizing the optimal temperature, precipitation, soil texture, and grower practices, such as tillage method and usage of cover crops, that contribute to the proliferation of certain pathogens, such as *Fusarium oxysporum*. The objective at hand is to adapt soil moisture models of various soil typologies for epidemiological modeling. Future research will attend to other edaphic conditions and cultural practices. Relevant data obtained from current and future projects will be used to construct models describing the epidemiology of various soil-dwelling fungi.

Understanding the effects of agricultural salinization on the incidence and severity of Fusarium wilt of tomato in California's Central Valley

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California's Central Valley produces 97% of all processing tomatoes in the United States. *Fusarium* wilt of tomato, caused by the soil borne fungal pathogen *Fusarium oxysporum* f. sp. *lycopersici* race 3 (FOL R3), is devastating processing tomato production statewide and has recently spread to Fresno County (producing 31% of CA tonnage). The disease appears severe in this county, causing greater than 50% yield loss within one to three years of detection in a given field. The Western San Joaquin Valley is experiencing issues with soil salinization as reductions in surface water allocations lead to increased use of saline groundwater. We initiated greenhouse trials to test the hypothesis that increasing soil salinity may enhance disease severity. Plants were set up in a split plot complete block design with two tomato cultivars – one susceptible and one tolerant to FOL R3 – saline and non-saline irrigation treatments, and inoculated and non-inoculated pathogen treatments. At 40 days post inoculation (DPI), incidence of FOL in inoculated plants was 100%. Extent of root colonization in both inoculated cultivars was 20% greater under saline irrigation than non-saline; extent of stem colonization did not differ between salt treatments (45–75% of stem colonized). At 40 DPI, root colonization for both cultivars was 43–50% higher under saline treatments compared to non-saline. At 90 DPI, fresh-weight was 23% lower in inoculated plants treated with salt compared to inoculated plants without salt. These results suggest that salinity may play a significant role in increasing FOL R3 colonization and enhancing disease severity, especially in tolerant cultivars.

Dollar spot control affected by bentgrass susceptibility and fungicide programming

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Creeping bentgrass (*Agrostis stolonifera* L.) susceptibility and fungicide timing affect dollar spot (caused by *Sclerotinia homoeocarpa* F.T. Bennett) control. This 3-yr field trial investigated fungicide timing effects on dollar spot on a high-susceptibility ('Independence') and low-susceptibility ('Declaration') cultivar. The trial was a $2 \times 8 \times 2$ factorial in a randomized complete block design with 5 blocks. Factors included bentgrass cultivar; initial fungicide timing [threshold-, calendar-, logistic regression model-, or growing degree day (GDD) model-based (20-30, 30-40, 40-50, 50-60, or 60-70 GDD; base 15°C and biofix 1 April)]; and subsequent fungicide timing [logistic regression model or disease threshold]. Response to treatments was limited in 2016 due to dollar spot suppression from fludioxonil applied to control anthracnose. Cultivar and subsequent fungicide timing had a greater influence than initial fungicide timing on dollar spot incidence during 2015 and 2017. Moreover, the logistic regression model- and disease threshold-based subsequent applications produced excellent control on 'Declaration' during 2015; whereas, only the logistic-regression model produced excellent control on 'Independence'. Three and four to five threshold-based applications were made to 'Declaration' during 2015 and 2017; whereas, six to seven and six to nine threshold-based applications were made to 'Independence' during 2015 and 2017, respectively.

An integrated approach towards management of canker diseases of almond in California

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Canker diseases are the main cause of tree mortality in almond orchards in California. Pruning wounds serve as the primary infection court for fungal pathogens although infection may also occur through bark cracks and mechanical injuries. Protection of wounds using fungicides and sealers has been broadly employed to control canker diseases of woody crops. Additionally, knowledge of pruning wound susceptibility according to the month of pruning has been used to determine the best timing for pruning. The goals of this study were to determine the duration as well as seasonal susceptibility of almond pruning wounds to various canker-causing fungi and assess the efficacy of several fungicides, biocontrol agents and wound sealants to protect pruning wounds. Field trials were conducted over two consecutive years in three commercial orchards in California. For the pruning wound susceptibility trials, fungi were inoculated on pruning wounds made between September and January and varying in age from 0- to 8-weeks-old. In the pruning wound protection trials twenty-one products were tested against seven canker-causing fungi. Preliminary results show that pruning wound susceptibility was highest for September pruning. Susceptibility of wounds decreased as wound age increased. Thiophanate-methyl and a *Trichoderma* biocontrol agent were the most efficacious products for protection of pruning wounds against the range of canker pathogens.

The impact of whole almond orchard recycling on prunus replant disease, basidiomycete, bacterial, and free living nematode populations

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The grinding and incorporating into soil of whole trees, during orchard removal, could provide a sustainable practice that enhances both air and soil quality. The objective of this project was to compare on-site grinding up and soil incorporation of whole trees with on-site burning and ash incorporation as a means of orchard removal. In 2009, each treatment was applied to seven replicate plots of an old stone fruit orchard in a randomized block design. An "Iron Wolf," a 45,454 kg rock-crusher, was used to grind up and incorporate standing tree rows of the old orchard to a soil depth of 30 cm. The grinding incorporated an estimated 67,364 kg of woody biomass per hectare. For the burn treatment, trees were pushed, burned, and the ash was spread throughout the plot. All replicate plots were replanted with almond trees in 2009. Significantly greater tree growth, yields, and soil nutrients were observed in the grind treatment. Significantly greater basidiomycete fruiting bodies and free-living nematodes were observed in the grind treatment. Evaluation of soil phospholipid fatty acid (PFLA) showed significant changes in soil microbiota after the application of wood chips. The wood chip treatment had significantly higher overall fungal and bacterial biomass, arbuscular mycorrhizae, saprophytes, actinomycetes, rhizobia, and protozoan populations. Cellulose degrading bacteria, *Chloroflexi* and *Cellulomonas* were significantly enriched in the wood chip plot. Over 8,000 hectares of old almond orchards have been ground and incorporated in the last three years.

Evaluation of copper-iodo-phosphonate on fire blight of pears

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Bacterial strains of *Erwinia amylovora* (Burrill) Winslow et al. resistant to streptomycin and tetracycline have prompted a search for effective control of the fire blight of apples and pears this bacterium causes. A recent nutrient formulation of copper-iodo-phosphonate (Cu-I-PO₃) shown to have bactericidal activity was compared to an orchard standard (OS) mixture of Agri-Mycin-17 (22.4% Streptomycin sulfate) and Mycoshield (31.5% Oxytetracycline Calcium) applied to a six year-old irrigated Bartlett pear orchard. Blossom Protect (*Aureobasidium pullulans*) and Buffer Protect (65.4% citric acid, disodium phosphate) were applied 17 March 2017 to the entire plot area. The first applications were made at 50% full bloom on 14 April 2017. Subsequent applications were based on weather for favorable infection. A total of nine applications of the OS treatment were made over the next two months compared with three applications of the Cu-I-PO₃ at either 900 or 1350 ml acre⁻¹ to coincide with the earliest OS applications. A complete randomized block design with four replicates was used. Data was recorded as percent 'hits' (infection sites) on 27 May and 23 July. The OS treatment had 13.2% 'hits' on 27 May and 63.5% on 23 July. In contrast, the low rate of Cu-I-PO₃ had 5.2 and 35.3% hits and the high rate had 2.3 and 42.0% on 27 May and 23 July, respectively. This data suggests that Cu-I-PO₃ may be a superior alternative to antibiotics for control of fire blight.

Optimizing genetics to manage Verticillium Wilt in tomatoes through traditional breeding and grafting: An elusive challenge

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The Ve locus in tomato confers resistance to race 1 strains but resistance to all non-race 1 strains of the *Verticillium dahliae* pathogen has eluded researchers for decades. The tomato line 7-1K, currently under selection at NC State University, has a high level of

resistance to *Verticillium* wilt symptoms. In a naturally infested 2017 field trial with 100% *Verticillium* incidence in a susceptible control, 7-1K had an incidence of 21% (n = 33) *Verticillium* related necrosis/chlorosis of the foliage. However; in the same trial a race 1 resistant scion Red Defender (RD) was grafted onto 7-1K rootstock, and incidence of foliar symptoms of the RD scion was 92%. These data indicate roots alone do not confer resistance, and that the foliage of non-grafted 7-1K is possibly tolerant to *Verticillium*. RD grafted on the vigorous rootstock Maxifort had an incidence rating of 89% but this combination generated the greatest yield of 70,285 lb/A in the absence of fumigation. Non-grafted RD on fumigated plots generated significantly less yields of 40,516 lb/A (P = 0.05). A range of other named and experimental rootstock selections had intermediate yields. Growth chamber assays showed all non-race 1 isolates were capable of colonizing 7-1K and 4 of 6 NC isolates colonized Aibou (a race 2 resistant rootstock) as assayed 6-wks after inoculation on isolations from upper stem tissue. These data suggest NC strains may be non-race 1 and non-race 2 and able to colonize these selections.

Genetic diversity of Grapevine leafroll-associated virus 4 in Washington State vineyards

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Among the recognized species of grapevine leafroll-associated viruses (GLRaVs), GLRaV-4 (genus *Ampelovirus*, family *Closteroviridae*) is made up of several strains that were previously considered as distinct GLRaVs. Among them, three GLRaV-4 strains (strain 4, strain 5 and strain 9) were previously reported from Washington State vineyards. Using a combination of high-throughput sequencing, Sanger sequencing and RACE methods, the genome size of GLRaV-4 strain 4 (isolate WAMR-4 from *Vitis vinifera* cv. Merlot), GLRaV-4 strain 5 (isolate WASB-5 from cv. Sauvignon Blanc) and GLRaV-4 strain 9 (isolate WALA-9 from cv. Lagrein) was determined to be 13,824 nucleotides (nt), 13,820 nt and 13,850 nt, respectively. The genome of GLRaV-4 strains 4, 5, and 9 characterized in this study contained seven open reading frames (ORFs) and the arrangement of these ORFs corresponded with the gene arrangement in GLRaV-4 strains reported previously from other grapevine-growing regions. ORF-by-ORF comparisons indicated that the three GLRaV-4 strains from Washington State vineyards were closely related to GLRaV-4 strains 4, 5, and 9 than to other GLRaV-4 strains reported from grapevine-growing regions outside the State. The WAMR-4 isolate contained two highly variable regions in the ORF1a, making it unique among the currently sequenced isolates of GLRaV-4 strain 4. Phylogenetic analysis showed segregation of GLRaV-4 isolates into distinct clades in a strain-specific manner. Recombination detection program (RDP4) identified putative recombination events between the genome sequences of GLRaV-4 strains. These results imply recombination as a potential means of evolution of distinct strains of GLRaV-4.

Characterization of phenotypic variation and genome aberrations in *Phytophthora ramorum*

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We have recently shown that the Sudden Oak pathogen *Phytophthora ramorum* undergoes irreversible phenotypic alterations accompanied by chromosomal aberrations when infecting trunks of mature oak trees (genus *Quercus*). In contrast, genomes and phenotypes of the pathogen derived from the foliage of California bay (*Umbellularia californica*) are usually stable. We define this phenomenon as host-induced phenotypic diversification (HIPD). *P. ramorum* also causes a severe foliar blight in some ornamental plants such as *Rhododendron* spp. and *Viburnum* spp., and isolates from these hosts occasionally show phenotypes resembling those from oak trunks that carry chromosomal aberrations. In this research, we cross-examined phenotypic variation and chromosomal aberrations in *P. ramorum* isolates from oak and non-oak hosts and substrates. The results suggest that HIPD comparable to that occurring in oak hosts occurs in non-oak environments such as in *Rhododendron* leaves. *Rhododendron* leaves are more easily available than mature oak stems and thus can potentially serve as a model host for the investigation of HIPD, the newly described plant-pathogen interaction.

Comparison of fungicide application methods to control postharvest diseases in apple

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The management of postharvest diseases of pome fruit mainly depends on the use of fungicides, which were traditionally applied via drench solutions at harvest. Thermo-fog and aerosolization are two new concepts of dry application in which a fungicide solution is heated and delivered as fine fog or mist inside the storage rooms. The efficacy and risks of the new methods have not been reported. Our study aimed to evaluate their efficacy compared to drench in controlling *Penicillium expansum*, *Botrytis cinerea* and *Neofabraea perennans*. Artificially inoculated fruits (cv. Fuji and Red Delicious) were treated with a mixture of thiabendazole + fludioxonil or pyrimethanil and each was applied via drenching, fogging and aerosolization. Fruit were stored at 0.5°C in a controlled atmosphere for up to 7 months. Disease incidence and severity were determined post-storage and fungicide residue levels on the fruit surface were evaluated. Our results indicate that drench is more effective than fogging or aerosolization in controlling the three pathogens. Moreover, the dry methods resulted in inconsistent residue levels throughout the storage rooms with areas showing significantly high and others significantly low fungicide residue levels on the fruit surface compared to drench. This study indicates potential increased risks of decay when applying fungicide dry and warrants improved applications and fruit set-up inside the storage rooms to enhance fungicide distribution.

Citrus dwarfing viroid infection results in reduced apical shoot growth in navel orange trees on trifoliolate rootstock

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Citrus dwarfing viroid (CDVd) infection of navel orange trees (*Citrus sinensis* (L.) Osb.) on trifoliolate orange (*Poncirus trifoliata* (L.) Raf.) rootstock has been previously reported to reduce canopy volume by approximately 50%. To understand how CDVd reduces canopy volume, a survey of navel orange tree growth was initiated in an experimental block planted in 1998 at the Lindcove Research and Extension Center (LREC) in the San Joaquin Valley, California. We first confirmed that 20 years after planting there was still a significant difference in tree height and canopy volume between CDVd-infected trees and non-infected controls. Since vegetative growth accounts for most of a tree's canopy volume, we then monitored vegetative apical shoot growth over two seasons in the CDVd-infected and non-infected control trees. We also surveyed the apical shoot growth of 34-year-old navel orange trees on flying dragon trifoliolate rootstock, commonly known to produce small-sized trees. We found that the net growth of the apical shoots of CDVd-infected trees was significantly lower (reduced by almost 30%, $p < 0.01$) than that of the control. We also determined that reduced apical shoot growth caused by CDVd was not significantly different from the effect of flying dragon rootstock on growth ($p = 0.8$). The findings of this study indicate that the effect of CDVd on apical shoot growth is likely to be the main factor determining the observed reduction in overall citrus tree canopy volume.

The National Clean Plant Network for Citrus: A model system for the prevention of citrus diseases

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Pathogen-tested, true-to-type propagative materials are the basis of a successful citrus industry. In 2005, three USDA agencies, APHIS, ARS, and NIFA, agreed that a national network is required to support programs generating, preserving and distributing clean propagative materials for specialty crops. The National Clean Plant Network (NCPN) was authorized by the US Congress in the 2008 Farm Bill and became permanent in 2014 with the mission of "providing high quality asexually propagated plant material free of target plant pathogens and pests that cause economic loss." Incorporation of citrus into NCPN began in 2007, and the charter for the NCPN-Citrus was adopted in 2010. NCPN-Citrus currently has 10 centers in 9 US states and territories, including CA, FL, AZ, TX, LA, AL, HI, MD, & PR. NCPN-Citrus centers introduce, test, maintain and distribute pathogen-tested citrus propagative materials for use by scientists, the general public and the multibillion dollar citrus industry. In a typical year, NCPN-Citrus centers conduct over 75,000 diagnostic tests, distribute over 600,000 units of clean materials, perform therapeutics on hundreds of accessions, maintain hundreds of foundation plantings of budwood sources, and foster various citrus extension programs. In conjunction with the other NCPN specialty crops, NCPN-Citrus develops outreach programs to educate and train industry personnel and the public on the importance of using pathogen-tested materials.

Taxonomy and phylogeny of cylindrocarpon-like fungi associated with root rot diseases of perennial fruit and nut crops in California

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Cylindrocarpon-like fungi are common and destructive root pathogens of grapevine and other economically important perennial crops around the world. In this study we surveyed orchards (almond, cherry, kiwi, olive, peach, and pistachio) and vineyards for plants exhibiting decline symptoms, including wilting, defoliation, and low vigor, between 2015 and 2017 in the Central Valley Region of California. In total, 39 representative isolates with cylindrocarpon-like asexual morphs were recovered in culture from symptomatic roots of diverse perennial crops. Detailed morphological observations of conidia paired with multi-locus sequence typing of three loci (ITS, beta-tubulin, and translation elongation factor 1-alpha) revealed eight previously described species including: *Campylocarpon fasciculare*, *Dactylonectria alcacerensis*, *D. macrodidyma*, *D. novozelandica*, *D. torresensis*, *Ilyonectria capensis*, *I. liriodendri*, *I. robusta*, and two new species that reside in the genera *Neonectria* and *Thelonectria*, respectively. Results from isolations of diseased plant roots revealed potential new host associations for all fungi in this study, except *Campylocarpon fasciculare* which is only known from grapevine, thus expanding our knowledge of possible novel host associations and illuminating new potential threats to the production and sustainability of economically important fruit and nut crops in California. This work lays the foundation for future studies on the epidemiology, pathogenicity, and host range of these important and destructive pathogens in California agroecosystems.

Soil and root microbiota of field-grown processing tomatoes at various spatial scales

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To assess the variation in microbial community composition along different spatial scales in agricultural settings, we profiled, using high-throughput Illumina sequencing of 16S rRNA and ITS amplicons, the bacterial and fungal communities in bulk soil, rhizosphere soil, and tomato crown tissue from plants being grown in six commercial processing tomato fields in the Sacramento Valley during three consecutive growing seasons. At the largest spatial scale, we found a strong effect of field location on the bacterial and fungal communities of both bulk and rhizosphere soil. At the smallest spatial scale, we observed a clear difference between bulk and rhizosphere soil communities. For the fungal community, 15 genera were more abundant in the rhizosphere: six of these, accounting for about half of the community, represented species that are known pathogens of tomato, including *Pyrenochaeta lycopersici*, *Plectosphaerella cucumerina*, *Fusarium oxysporum*, *Colletotrichum coccodes*, *Fusarium solani*, and *Rhizoctonia solani*. Together with

Verticillium dahliae, these same species were also found in tomato crown tissues, accounting for 39% of the community. The discovery of *P. cucumerina* was unexpected because it has not previously been described as a tomato pathogen of concern in the US. We successfully isolated *P. cucumerina* from these same soil samples and were able to produce symptoms on tomato roots. Our study demonstrates the utility of high-throughput based microbial community analysis in uncovering hidden pathogen-host relationships while providing a scale-dependent understanding of the plant microbiome in agroecosystems.

Biological products update by Isagro USA, Inc.

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Isagro USA (<http://www.isagro-usa.com/>) has been focused on developing and growing its bio-solutions portfolio designed to serve growers seeking to meet consumer demand for more natural and organic food production. The proprietary portfolio has been expanding with products such as DOMINUS®, a broad-spectrum soil biofumigant that effectively controls important soil-borne fungi, nematodes, weeds, and insects (pending USDA NOP approval, 2018); BIO-TAM® 2.0, an OMRI-listed biorational fungicide on fruit and vegetable crops; KENTAN® DF, an OMRI-listed copper hydroxide bactericide/fungicide with a higher degree of rainfastness; and TAEGR0® 2, an OMRI-listed biofungicide that provides broad-spectrum protection of major soil-borne and foliar diseases. Current data will be presented.

Chasmothecia sampling as a pre-season indicator of QoI fungicide resistance in *Erysiphe necator*

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Control failures of grape powdery mildew caused by *Erysiphe necator* after the use of quinone outside inhibitor (QoI; FRAC 11) fungicides have been reported throughout OR, WA, and CA. Quick, cost-effective methods to detect fungicide resistance in *E. necator* are necessary to reduce the selection for resistant isolates, mitigate ineffective fungicide applications, and ease labor requirements. A rapid sampling technique for chasmothecia (syn. cleistothecia) overwintering on grapevine bark is being developed to provide managers with a pre-season assessment of the risk of fungicide resistance for the growing season following sampling. The wild-type and resistant genotypes can be determined from a single chasmothecium in a background of 0.2 g of dry bark using a bead beater-based DNA extraction process and analyzed using a competitive TaqMan qPCR assay for the G143A mutation in the *cytb* gene - previously shown to be in 100% agreement with QoI resistance bioassays. Leaves and bark samples were collected in a linear stratified sampling pattern from 12 commercial vineyards in the Willamette Valley of OR during Winter 2017–18. Results will be compared to Spring 2018 samples collected using in-season rapid sampling techniques (e.g. swab conidiospores from worker gloves after conducting canopy management practices) to determine the predictive ability of chasmothecia sampling, including an examination of spatial and temporal sampling density requirements.

Assessment of *Fusarium oxysporum* f. sp. *vasinfectum* race 4 as a seedling pathogen to cotton, common bean, and soybean using a rolled towel assay

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Fusarium oxysporum is an important fungal pathogen that causes wilt disease to many economically important crops such as cotton, common bean, and soybean. *Fusarium oxysporum* f. sp. *vasinfectum* (FOV) race 4 is an aggressive wilt pathogen towards cotton that can also cause seedling mortality. FOV race 4 has negatively impacted cotton in California since 2001, and in 2017 it was reported in Texas. As crop rotation is common practice, a better assessment of the pathogenicity of FOV race 4 on various plant hosts can help to determine how the fungus persists in the absence of its primary host. The objective of this study was to assess the reaction of cotton, common bean, and soybean to seedling infection when inoculated with four isolates of FOV race 4 using a rolled towel assay. Seeds for each host were inoculated with 100 µl of a 1×10⁶ conidia/ml suspension. At 14 days seedlings were rated using a disease severity index (DSI). All hosts were significantly impacted by all isolates of FOV race 4 when compared to the non-inoculated controls ($P < 0.05$). There was a significant difference for isolate and host ($P < 0.001$), with cotton having the highest DSI among hosts. The results from this study suggest that FOV race 4 can persist in the field in the absence of cotton on other hosts and can also negatively impact these hosts. Further testing of FOV race 4 and their affect on other hosts commonly used in crop rotation with cotton should be explored.

Genotypic and phenotypic characterization of pathogenic *Streptomyces* spp. on potato and turnip in Michigan

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Common scab of potato is caused by *Streptomyces* species. There are over 400 species of *Streptomyces*, however few of them are plant pathogenic. To date, at least 13 different species of *Streptomyces* can infect and cause disease on potato and other crops. *Streptomyces scabies* is the most common and widely distributed pathogenic species among the streptomycetes. For this study, we characterized plant-pathogenic *Streptomyces* spp. associated with common scab infected potato and turnip tubers harvested in Michigan. A total of 51 pathogenic isolates from pitted and surface scab lesions were recovered from potato and turnip tubers. The isolates were identified by

morphological and molecular methods. Preliminary results showed that 48 isolates causing pitted and surface lesion have genes encoding for thaxtomin synthase (*txtA* and *txtAB*), tomatinase (*tomA*) and a necrosis protein (*nec1*). Moreover, preliminary results of sequence analysis of the 16S rRNA gene indicated that 10 of the isolates were similar to *S. scabies*. Additionally, sequence analyses of the thaxtomin synthase, tomatinase, necrosis protein and three housekeeping genes (*rpoB*, *recA* and *atpD*) were performed.

Investigating the role of *Colletotrichum karstii* in twig and branch dieback of citrus in California

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Colletotrichum Dieback (CD) is an emerging disease of Clementine mandarin and navel oranges in California. Common symptoms include twig and shoot dieback, and in severe cases, branch dieback of trees. The most characteristic symptoms are bark splitting and gumming on young shoots. Field surveys of ten citrus orchards were conducted in the San Joaquin Valley region between 2014 and 2017 in order to determine the occurrence of *Colletotrichum* species associated with CD in California. Morphological identification along with phylogenetic analysis of three gene regions (ITS1-5.8S-ITS2, b-tubulin, and GADPH) confirmed the presence of two distinct *Colletotrichum* species associated with CD in the San Joaquin Valley: *C. gloeosporioides* and *C. karstii*. Both *C. karstii* and *C. gloeosporioides* caused internal vascular necrosis when inoculated into healthy Clementine trees, confirming both species as pathogenic. Ongoing spore trapping studies show the majority of *Colletotrichum* spores are present during the spring months. This study establishes CD as an emerging disease of citrus in California, particularly in the San Joaquin Valley, and confirms *C. karstii* as a new pathogen of citrus in California.

Infection of walnut by Botryosphaeriaceae and implications in disease control management

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Walnut cultivation occupies about 450,000 acres in California. Walnuts are considered one of the very health-promoting nuts due to their ω-fatty acids. A Botryosphaeria canker and blight disease has resulted in reduced yields in the last decade or so. Ten Bot/ceae species were identified from cankered and blighted tissues. Disease symptoms on spurs and fruit develop in mid- to late summer, although under favorable conditions, blighted shoots with cankers can show in late spring. The Bot/ceae fungi can infect any tissues wounded by pruning, sunburn, freeze, hail, walnut scale, and woodpeckers. Specifically, pruning wounds remained susceptible to infection for at least 4 months, and this is because the pathogens can establish in the pith of shoots. Starting from May to about mid-September, developing fruit on trees were inoculated with a spore suspension of *Neofusicoccum parvum* and kept on the trees until mid-September when they were harvested and evaluated for infection (blight) and kernel mold. Fruit inoculations indicated that the pathogen caused latent infections throughout the season and symptoms developed after the fruit had matured. Fruit infection moved into the stem and then in the sustaining spur, killing fruiting buds. Protection of fruit with fungicides (i.e. strobilurins, carboxamides, and mixtures) significantly reduced the disease and improved yields. An estimation of crop losses due to Botryosphaeria canker and blight will also be presented.

Grapevine leafroll-associated virus 3 is present as genetically distinct strains in Washington State vineyards

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Grapevine leafroll-associated virus 3 (GLRaV-3, genus *Ampelovirus*, family *Closteroviridae*) is the most insidious and damaging viral pathogen of wine grapes (*Vitis vinifera*) worldwide. This project was initiated to explore the genetic diversity of GLRaV-3 in vineyards across distinct wine grape-growing regions in Washington State. A total of 395 samples were collected randomly over several seasons from eight red-berried and four white-berried cultivars showing and/or suspected for grapevine leafroll disease (GLD) symptoms from ten commercial vineyards. Samples were processed and tested by RT-PCR for the presence of GLRaV-3 using primers specific to a portion of the heat shock protein 70 homologue (Hsp70h) gene encoded by the virus. The approximately 580 base pair (bp) region of the Hsp70h gene amplified in the PCR assays was cloned and sequenced, and the derived virus sequences were aligned with the corresponding sequences of GLRaV-3 reported from other grapevine-growing regions. A global phylogenetic analysis of the Hsp70h gene sequences revealed the presence of genetically diverse populations of GLRaV-3 in vineyards, with the majority of isolates aligning with variant group I compared to other variant groups of the virus. Further analysis showed that the majority of virus-positive samples from individual vines contained a single variant and nearly 20% of the samples were found to have sequences belonging to two or more variant groups of GLRaV-3. The results provided a baseline to further examine genetic diversity of GLRaV-3 for applications in improved diagnosis of the virus in grapevine planting materials and leafroll disease management.

Aphid resistance screening using high resolution melting marker and rapid DNA extraction protocol

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Screening seedlings for valuable agricultural traits with molecular markers can be costly and time consuming. For this reason we tested a rapid DNA extraction protocol to screen 900 black raspberry (*Rubus occidentalis*) seedlings for aphid resistance. We used a high-resolution melting (HRM) marker, S99_122915_HRM, that indicates presence/absence of aphid resistance, that has been mapped to *Rubus* linkage group 6. Using this protocol, we screened 900 samples with the marker within 5 days and at a reduced cost compared with our standard DNA extraction protocol. Marker S99_122915_HRM amplifies a fragment that is present in resistant individuals and absent in susceptible individuals. Controls that had previously been validated with this marker were included in the screen. The resistant parents for the screened progeny plants were selected based on resistance source (Ontario, Canada, Maine, or Michigan), and the degree of overall heterozygosity. The selection of susceptible parents for crossing was hindered by the high level of homozygosity in the available cultivars. A search for susceptible parents with potentially higher heterozygosity was performed by assessing wild genotypes susceptible to aphids. This search was performed using genotyping by sequencing (GBS) and giving wild genotypes quality scores based on quantity of heterozygous SNP calls. HRM analysis indicated 467 resistant seedlings to be planted for further evaluation.

***Aphanomyces euteiches* detection and in-field location in Montana**

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The discovery of *Aphanomyces euteiches* in northeastern Montana fields in 2016 has prompted further exploration into the in-field distribution of the pathogen. *Aphanomyces* causes root rot on peas and lentils. In 2017, two fields where the pathogen was previously discovered were surveyed. Soil samples were collected from low-lying areas in the field and the field entrance in 100 m transects. The field entrance was targeted as the pathogen is soil borne and spread by human movement and on equipment. Low spots allow water collection, ideal conditions for the swimming biflagellate zoospores. At 10 m intervals along the 100 m transect, five 6-inch soil cores were collected at random and combined to make a composite sample for each 10 m location. Soil samples from both fields were assessed in the greenhouse for pathogen presence using a bioassay baiting technique with a susceptible pea variety, DS Admiral. Plant roots were evaluated for disease severity using a scale of 0-5. There was weak evidence ($p = 0.07$) of a difference in root rot severity along the 100 m field entrance transect from one field, and root rot severity was too low in the other field to detect a difference. Isolates of *A. euteiches* were recovered from plant roots using MBV selective media and transferred to PDA for morphological identification. Molecular characterization is ongoing. Soil will be collected from additional fields in 2018. This project will measure the abundance and pathogenicity of *A. euteiches* in Montana, provide best sampling practices for detecting the oomycete, and provide information on pathogen diversity that can be used in disease management strategies.

Comparison of water-mold populations in Ohio using traditional isolation methods and an amplicon based metagenomics approach

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Sustainable disease management requires an understanding of the pathogen communities present in the field. This study investigates the diversity and relative abundance of water-mold species using traditional isolation methods (TIM) and an amplicon-based metagenomics approach (ABMA). A total of six locations across Ohio were selected for this study where seedlings and the corresponding rhizospheric soil were collected fourteen days after planting (dap). Isolates recovered from symptomatic seedlings on selective media were identified using Sanger sequencing of the full ITS region while the Illumina Mi-Seq was used to sequence water-molds present in the soil using the ITS1 region. With TIM, 2 species of *Phytophthora*, 15 *Pythium* and 3 *Phytophthora* were recovered. In contrast, the ABMA had 7 species of *Phytophthora*, 28 *Pythium*, and 3 *Phytophthora*. Additionally, the *Phytophthora* species *P. vexans*, *P. chamaecephalon* and *P. mercuriale*, were found in very low abundance in both methods. The ABMA identified a greater diversity of *Pythium* and *Phytophthora* spp. although this may not represent the most pathogenic species in the field. The use of selective media may have contributed to the lower species diversity when using TIM. Also, water-mold species known to be slow growers in culturing media contributed to the greater isolation of some *Pythium* spp. compared to *Phytophthora* spp. Furthermore, sampling at 14 dap recovered species that were active under those environmental conditions only, limiting the diversity when using TIM. Further experiments should include ABMA from infected plant tissue as well as the rhizospheric soil to assess whether ABMA results in a better recovery and characterization of water-molds compared to TIM.

Identification and pathogenicity of fungal species associated with crown and root rot diseases of pistachio in California

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In the last three years, we surveyed the main pistachio growing counties in California to assess the causes of various declines affecting pistachio trees. Soil borne diseases were detected and included crown and root rots. Symptoms comprised trees with chlorotic foliage, wilting, defoliation, gumming of the trunk, crown and root rots and eventually tree death. Isolation work and taxonomic studies revealed the occurrence in pistachio of several *Phytophthora* species including *Phytophthora niederhauserii*, *P. cinnamomi* and *P. taxon walnut*. The fungus *Macrophomina phaseolina* was also isolated from declining young pistachio rootstocks. Pathogenicity of these various soil-borne fungi was assessed using mycelium plug inoculations in one-year-old potted UCB-1 rootstocks maintained in a greenhouse.

Results showed that all the various fungi could colonize and damage the vascular tissues of UCB-1 rootstocks, causing crown rot. A second pathogenicity test was conducted by inoculating the roots of four-month-old potted plants including UCB-1 clonal and Platinum rootstocks using zoospore suspension for *Phytophthora* species and microsclerotia suspension for *M. phaseolina*. All fungi tested were able to cause root and crown rots in both UCB-1 clonal and Platinum rootstocks resulting in complete wilting and death of plants after three weeks. Platinum rootstocks appeared more tolerant to *Phytophthora* pathogens than clonal UCB-1 rootstocks whereas *M. phaseolina* affected both rootstocks equally, resulting in the killing of all inoculated plants. Additional experiments are on-going to investigate the susceptibility of the main pistachio rootstocks including UCB-1, PGI and Platinum to all these various soil-borne pathogens.

Epidemiology of soilborne inoculum of *Phytophthora ramorum* in nurseries

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Soilborne inoculum has been proposed to be an important contributor to the local persistence of the sudden oak death pathogen *Phytophthora ramorum*, however current research has only assessed pathogen survival. Two additional aspects of the disease cycle were investigated at the National Ornamental Research Site at Dominican University of California: sporulation capacity post incubated in soil, and the infection of leaf baits at the soil surface. Rhododendron leaf disk inoculum was buried in soil at 5 and 15 cm. Periodically over the course of a year, we baited subplots with non-infested leaf disks at the soil surface and recovered inoculum for survival and sporulation assessment. Recovery of *P. ramorum* remained generally high; sporangia production remained greater for inoculum buried at 15 cm than at 5 cm, but declined for inoculum at both depths. With the onset of cooler temperatures in autumn we observed a moderate increase in sporangia production, corresponding to the time period with greatest incidence of recovery from the bait disks. Laboratory experiments confirmed chilling results in an increase in sporulation capacity. While sporulation capacity was greater at deeper depths, the ability of spores to reach the soil surface was reduced relative to the shallow depth. Soil inoculum may initiate new disease cycles, with greatest risk in the months following the onset of cooler temperatures.

Genetic diversity of *Rhizobium leguminosarum* and *Mesorhizobium ciceri* from pea and chickpea fields in the Palouse

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Legumes support symbiotic associations with rhizobacteria that result in biological nitrogen fixation that serves as a nitrogen fertilizer for the legume host and subsequent rotation crops. In some cases, symbiosis genes are clustered and in other distinct cases, they are dispersed. Even though it is evident symbiosis is important globally, only a limited number of *Rhizobium* genomes have been sequenced. The objective of this research is to examine genetic diversity of *Rhizobium leguminosarum* and *Mesorhizobium ciceri* isolates collected from peas and chickpeas grown in the Palouse region of Washington and Idaho. DNA was extracted from each isolate and amplified by PCR. Genetic relationships were determined by DNA sequence analysis across several genes including, *atpD* (ATP synthase), *nodC* and *nifH* (nodulation and nitrogen fixation), *recA* (DNA repair), *GSI* and *GSII* (glutamine synthetase), and phylogenetic trees were constructed. A total of 75 isolates of *R. leguminosarum* were examined from five different pea fields in addition to 83 isolates of *M. ciceri*, which included 1 commercial isolate and 82 isolates collected from four chickpea fields. Phylogenies based on multi-locus sequencing suggest that isolates of *M. ciceri* from chickpea are more diverse than isolates of *R. leguminosarum* from pea. These differences may be attributed in part to the recent use of different commercial *M. ciceri* inoculants in Washington and Idaho. This collection of genetically characterized isolates will be used to screen chickpeas and peas for the ability to form symbiotic associations with rhizobacteria.

Types and roles of melanin in the grapevine phytopathogenic fungus *Lasiodiplodia exigua*

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Melanin is a pigment produced by the oxidation of phenolic and indole compounds, and in some fungi is essential for pathogenicity. *Lasiodiplodia* spp. are considered among the most aggressive pathogens in grapevine, causing dieback. A common characteristic of this genus is the presence of melanized spores and mycelium. To characterize the types and roles of melanin in *L. exigua* str. UCD256Ma, specific melanin inhibitors were used. It was found that this fungus has the ability to synthesize: DOPA-melanin, DHN-melanin, and pyromelanin. DOPA-melanin was found to be related to the production of aerial mycelium and to help with the protection against enzymatic lysis and oxidative stress. DHN-melanin is involved in the ramification of the mycelium in the lack of nutrients, and pyromelanin is associated with hyphae development. On another hand, tyrosine is directed to the production of DOPA-melanin and the fungus used it as carbon and nitrogen source; thus DOPA-melanin is the principal melanin produced by the fungus. Last, a histological study of grapevines cv. Cabernet Sauvignon inoculated with *L. exigua*, showed the presence of melanized hyphae mainly in the pith and parenchymatic cells of the plant, but not in the vascular bundles, indicating that the melanization in the mycelium increases with the progress of colonization of the plant. Thus, synthesis of melanin inside the plant might help the fungus to protect itself from the mechanisms of defense of the plant.

Sensitivities of *Botrytis cinerea* pear isolates to registered and new fungicides

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Grey mold, caused by *Botrytis cinerea*, is a common postharvest disease in pear. Only few fungicides are registered for pre and postharvest application to manage postharvest rots. We tested the sensitivity of *B. cinerea* against four fungicides with different modes of action (Cyprodinil, Triflumizole, Dodine, and Iprodione) using mycelial growth inhibition assays. Twenty-one fungal isolates collected from pear orchards in Medford, OR in 2017 were grown for three days on PDA amended with fungicides at 0, 0.01, 0.1, 1, and 10 µg/mL. Dodine and Cyprodinil were also tested at 100 µg/mL and at maximum labeled field rates. Cross sectional colony diameter was measured for each sample plate. The effective concentration to reduce radial growth by 50% (EC50) was calculated for each pathogen-fungicide combination. The EC50 for isolates against Cyprodinil, Triflumizole, Dodine, and Iprodione ranged from 0.95 to 360, 0.38 to 1.43, 12.34 to 195.65, 0.57 to 1.13 µg/ml respectively. Except for Cyprodinil, the tested isolates were sensitive within labeled field rates. Two isolates (10%) were not sensitive against Cyprodinil at the maximum labeled field rates and EC50 for 85% isolates were greater than 5 µg/ml. These results suggested a possibility of resistant *B. cinerea* population to Cyprodinil, and inclusion of three additional groups of fungicides in a rotation program for managing postharvest rots on pears.

Phomopsis stem canker: An emerging threat to hazelnut production in British Columbia

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Hazelnut (*Corylus avellana*) production in British Columbia is concentrated in the Southeastern regions in the Fraser Valley. Since 2001, hazelnut orchards have been affected by eastern filbert blight (EFB), caused by *Anisogramma anomala*. Because many of the orchards with older varieties are in decline, affected orchards are being replaced with EFB-resistant varieties. A preliminary survey conducted in 2017 shows that many of the young orchards planted with EFB-resistant varieties, 'Jefferson', 'Theta' and 'Yam Hill', are now being subjected to a new threat associated with a *Phomopsis* sp. Affected orchards of 6- to 8-year-old hazelnut show symptoms of defoliation and dieback of branches, mainly due to enlarging cankers on stems and trunks. These cankers appear as brown to dark-brown lesions with light brown margins. As the cankers mature, the pathogen produces tiny, dark coloured pycnidia and pycniospores typical of *Phomopsis* sp. In some cases, tissues underneath the cankers deteriorate, resulting in sunken lesions with cracked margins. Infected stems display a 'V-shaped' brown discolouration of the internal tissues beneath the lesions. *Phomopsis* sp. isolated from the canker tissues was confirmed by morphology and DNA sequencing. Although *Phomopsis revellens* Hohn. associated with a canker disease on filbert was documented from Sumas Prairie in 1939, no such disease has been reported from hazelnut orchards since their establishment in the Fraser Valley. However, stem and twig canker caused by a *Phomopsis* sp. has been reported from southern Chile in 2014. Currently, studies have been initiated to confirm the pathogenicity, species identification and disease epidemiology of the *Phomopsis* sp.

Disarming *Macrophomina phaseolina*: Could Arabidopsis help discover a means to circumvent a global destroyer of crop plants?

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Dubbed as a "global destroyer of crops", the necrotrophic soilborne fungus *Macrophomina phaseolina* (*Mp*) causes diseases such as charcoal rot, canker, damping off and wilt on more than 500 plant species including many economically important crops. Host plants stressed by inadequate water availability in rain-fed cropping systems are especially susceptible to *Mp* infection. Molecular mechanisms of host defenses against this pathogen are poorly understood. We established interactions between *Mp* and *Arabidopsis thaliana* (*Arabidopsis*) as a model to quantitatively assess host factors affecting the outcome of *Mp* infections. Using agar plate-based infection assays we quantified *Mp* biomass growth in roots of different *Arabidopsis* genotypes. By comparative profiling of host transcriptomes in roots of the wild-type *Arabidopsis* accession Col-0, with and without *Mp* infection, we uncovered expression changes in hundreds of genes potentially contributing to *Mp* defense mechanisms or putative effector targeting schemes. Our results clearly indicate *Mp*/*Arabidopsis* interactions to be a useful model pathosystem, which allows for the application of the vast genomics-related resources of this versatile model plant to the systematic investigation of previously understudied host defenses against a major broad-host spectrum plant pathogen.

Pathogenic *Rhodococcus* associated with previously unreported woody and herbaceous plant hosts

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The Gram-positive bacterium *Rhodococcus* causes leafy gall disease on over 100 plant taxa, resulting in substantial losses for growers of ornamental plants since there is no cure for infected plants. Phytopathogenic *Rhodococcus* isolates have been classified into at least 13 distinct clades. Since 2007, the Oregon State University Plant Clinic has recovered pathogenic *Rhodococcus* in 28 plant species not previously reported as hosts. Plants were received from 12 states and included: *Agave potatorum*, *Angelonia* × 'Actors White', *Aruncus aethusifolius*, *Aruncus* × 'Misty Lace', *Calendula* sp., *Campanula punctata*, *Choisya ternata*, *Coreopsis* × 'Jive' and 'Crème Brûlée', *C. verticillata*, *Echinacea purpurea*, *Erodium reichardii*, *Gomphrena globosa*, *Lagerstroemia* × 'Infinitini Brite Pink', *L. indica*,

Lamium maculatum, *Physalis alkekengi*, *Physocarpus* sp., *Scabiosa columbaria*, *Sedum acre*, *S. lineare*, *S. reflexum*, *Sorbaria sorbifolia*, *Stokesia laevis*, *Sutera cordata*, *Veronica* × ‘Magic Enchanted’, *V. officinalis*, *V. umbrosa*, and *V. whitleyi*. This increase in the number of plant species affected likely reflects the volatility of the market regarding new introductions, as well as improved detection methods. Nurseries with chronic *Rhodococcus* problems may have resident populations that generate new lineages. Stringent sanitation is essential to limit losses in nurseries.

Analysis of global populations of *Phytophthora cinnamomi* suggests presence of two dominant clonal lineages and evidence of sex in Southeast Asia

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Phytophthora cinnamomi is a heterothallic, broad host range plant pathogen causing dieback and root rots of more than 3000 plant species. Several independent studies have suggested the existence of clonal lineages, primarily of the A2 mating type, and rarely sexual reproduction. However, a rigorous study of population diversity at a global scale is currently lacking. We analyzed 197 isolates of *P. cinnamomi* sampled from 11 countries. Genotyping by sequencing was performed using two restriction enzymes (PstI and MspI) and sequenced on the Illumina HiSeq 3000 platform. Raw reads were mapped to the *P. cinnamomi* reference genome using bowtie2 and variants were called with the GATK HaplotypeCaller. We tested the hypothesis of clonal reproduction in *P. cinnamomi* populations. A neighbor-joining tree based on 1,027 variants indicated that populations of *P. cinnamomi* from Southeast Asia (Taiwan & Vietnam) consisting of A1 and A2 mating type isolates were highly diverse whereas most isolates from the remaining countries formed two distinct clonal lineages of the A2 mating type. A1 mating type isolates from Papua New Guinea, South Africa and Australia grouped within the diverse Taiwanese and Vietnamese populations which were inferred to be partially sexual based on the index of association. This suggests that Southeast Asia might be a candidate center of origin for *P. cinnamomi* as speculated previously. These results provide novel insights into the existence of both mating types in Southeast Asia and probably sexual reproduction that could potentially give rise to novel aggressive genotypes or lineages.

Comparison of fungicides for control of *Rhizoctonia solani* in sugarbeets

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Rhizoctonia root and crown rot (*Rhizoctonia solani* J. G. Kühn) is a major disease in a variety of crops around the world. Sugarbeets are very susceptible, and yield losses often range from 2% to 60% on average. As the U.S. is the fifth largest producer of sugar in the world, the potential effect of the losses can be tremendous. To compare fungicides for control of Rhizoctonia, a meta-analysis was performed to study the effects of pre-emergent treatments (in-furrow and seed applications) and post-emergent treatments (banded applications) as methods of controlling the disease in sugarbeet. Trials were conducted at the Southern Agricultural Research Center in Huntley, Montana from 2013 to 2017. In each year there was a high negative correlation between disease index and yield ($r = -0.85$ on average). Repeated measures analyses showed that specific post-emergent treatments promoted significantly higher stand counts, lower disease index, and higher yield and recoverable sugar content. Pre-emergent treatments as a whole had less of an effect, but there were combinations of pre- and post-emergent treatments that were associated with lower disease.

Characterization of Glutathione S-transferase (GST) and Superoxide dismutase (SOD) from the potato cyst nematode *Globodera pallida*

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The pale cyst nematode (PCN), *Globodera pallida*, is one of the most damaging plant parasitic nematodes of the potato and can cause multi-million dollar crop losses annually. *G. pallida* is a sedentary endoparasitic biotroph that penetrates roots of its host as a second stage juvenile (J2). After intracellular migration, J2s establishes a feeding site, if the female remains sedentary. Once fertilized, the female produces eggs and her body becomes a cyst. Encysted eggs can survive in soil for more than 20 years and are impervious to a wide variety of environments. During infection, J2s use a combination of physical (stylet) and chemical (effectors) methods to migrate through plant roots. Effectors, released through the nematode stylet may suppress plant defense responses or may induce a hypersensitive response (HR). HR triggers production of the reactive oxygen species (ROS) which create a cytotoxic environment for the nematode. Nematode may protect itself from ROS through activation of detoxification and antioxidant pathways. Glutathione S-transferases (GST) are detoxification enzymes that may play a role in the excretion of endogenously and exogenously produced toxic compounds, such as ROS. Superoxide dismutase (SOD) is an antioxidant enzyme that has a role in neutralization of the ROS. The main objectives of this research is to isolate and localize the expression of the *Gp-gst* and *Gp-Mn-sod* encoding genes from *G. pallida*. Both genes were amplified from the J2 cDNA, and cloned into the pCR[™]8/GW/TOPO vector. *In situ* hybridization was conducted to localize the transcript of the *Gp-gst* and *Gp-Mn-sod* in the *G. pallida* J2s. Putative role of these enzymes could give insights into possible control managements for *G. pallida*.

A host-specific RxLR effector of *Phytophthora palmivora* contributes to virulence on cacao

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Phytophthora palmivora is an oomycete plant pathogen with a wide range of hosts, including many economically important ones such as papaya and cacao. Its host specificity is largely unknown, but exists. For example, the cacao isolate does not infect papaya, and vice versa. The *P. palmivora* genome encodes a large set of effector proteins, including the RxLR family, which are believed to play key roles in pathogenicity. From the published genome and transcriptome data, we identified an RxLR effector designated PpalRxLR1 as highly induced during infection of cacao by a cacao isolate, but absent in the papaya isolate. PCR and RT-qPCR confirmed these findings. Three near identical copies were found in the genome assembly. A single guide RNA targeting all three was used to generate mutants via CRISPR/Cas9 gene editing. We obtained three single zoospore derived mutants with PpalRxLR1 either completely or partially mutated. The mutants exhibited reduced pathogenicity on cacao compared with the wild-type strain. PpalRxLR1a was predicted to have a nuclear localization signal using Localizer, and its nuclear localization was confirmed by transiently expressing the mature protein of PpalRxLR1a fused with DsRed in *Nicotiana benthamiana*. Altogether, these data suggest that PpalRxLR1 mediates *P. palmivora* pathogenicity likely by manipulating host gene expression. Its presence in the cacao but not papaya isolate suggests that it may play a role in host jump of *P. palmivora*.

Less common fungi isolated from grapevines in British Columbia and their role in the trunk disease complex

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Grapevine trunk diseases (GTD) are responsible for important economic losses and are caused by a wide range of taxonomical unrelated fungi, which primarily belong to the Botryosphaeriaceae, Diatrypaceae, Nectriaceae, and Togniniaceae families. Field surveys conducted in British Columbia (BC) between 2010 and 2015 found several fungi less commonly associated with GTD symptoms. Morphological studies along with multi-locus phylogenetic analyses identified six different *Valsa/Cytospora* spp., two *Neofabraea* spp., *Truncatella angustata*, and a *Pestalotiopsis* sp., all isolated from grapevine cankers. In order to determine the role that these fungi play in the GTD complex, pathogenicity tests were conducted in potted vines in a greenhouse. Actively growing mycelium plugs from two isolates of each of the ten species were used to inoculate one year-old dormant Chardonnay and Merlot rooted cuttings. Non-colonized agar plugs were used as negative control. Ten cuttings per treatment and per cultivar were used and plants were maintained under controlled temperature and relative humidity conditions. Six months after inoculation the extent of vascular discoloration was measured and re-isolations were conducted to fulfill Koch’s postulates. Results showed all fungal spp. to be able to cause vascular discoloration in both cultivars; however, virulence varied significantly depending on the species. *Valsa/Cytospora* spp. were shown to be the most virulent followed by *Truncatella angustata*, *Neofabraea* spp. and *Pestalotiopsis* sp. This study expands the knowledge on the GTD etiology in BC, which will contribute to the development of best management strategies against these diseases.

Characterization of *Xanthomonas arboricola* pv. *corylina* causing bacterial blight of hazelnut

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Bacterial blight (*Xanthomonas arboricola* pv. *corylina* (*Xac*)) of hazelnut (*Corylus avellana* L.) was described first in Oregon in 1915 and is now recognized as a damaging disease of young hazelnut trees worldwide. Thousands of acres of hazelnut cultivars that are resistant to a canker disease called eastern filbert blight (*Anisogramma anomala*) are being planted in the Willamette Valley of Oregon, where 99% of the US hazelnut crop is grown. Stressed hazelnuts are more susceptible to bacterial blight in conditions such as planting on marginal sites. There has been an increased incidence of bacterial blight in young hazelnut plantings, but no research on the disease has been done in Oregon since the 1970’s. We characterized bacterial isolates from symptomatic tissues for growth on semi-selective media, nutrient utilization profiles with Biolog Gen II, quinate metabolism, copper resistance, hypersensitive response (HR) on tobacco, and pathogenicity on hazelnut. Additionally, isolates were identified with a duplex PCR assay (*ftsX* and *qumA*), 16S rRNA sequence, and multilocus sequence analysis (MLSA) using *rpoD* and *gyrB*. Pathogenic isolates were identified as *Xac* using morphological, biochemical, and molecular assays. With MLSA, *Xac* isolates separated into two clades, one clade with the type strain and a second clade previously described with isolates from Poland. Thus, the phylogenetic diversity of *Xac* observed in other countries also is present in Oregon. Future studies will evaluate sensitivity of the newly released OSU hazelnut cultivars to bacterial blight with *Xac* isolates from the two clades.

***Sclerotinia sclerotiorum* secretes a small peptide that specifically interacts with plant polygalacturonase-inhibiting protein (PGIP)**

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The necrotrophic pathogen *Sclerotinia sclerotiorum* infects more than 400 plant species including many economically important crops. It causes various diseases called white mold or Sclerotinia stem rot, with rotting as a prominent symptom due to maceration of host tissue. *S. sclerotiorum* produces a number cell wall-degrading enzyme including polygalacturonases (PGs) that degrade pectin in the plant cell wall. To protect plant cells from fungal PG activities, plants have evolved to produce polygalacturonase-inhibiting proteins (PGIPs) that bind to pathogen’s PGs and mitigate PG activity. There have been no reports of pathogens’ ability to counteract plant

PGIPs. Here we show *S. sclerotiorum* secretes a small effector peptide (E1) that specifically interacts with plant PGIP. *E1* is significantly up-regulated during early stage of infection, and deletion of *E1* resulted in a significant reduction in virulence. Through yeast two-hybrid (Y2H), co-immunoprecipitation (co-IP) and bimolecular fluorescence complementation (BiFC) assays, we demonstrate that E1 specifically interacts with AtPGIP1 of *Arabidopsis thaliana*. E1 purified from cultures of *E1*-expressing yeast cells has no PG enzyme activity, but binds to AtPGIP1 and reduces effectiveness of PGIP in PG activity assays. Our results show that *S. sclerotiorum* secretes a special effector that interferes with plant PGIP, reducing PGIP inhibitory effect and enhancing PG activity in supporting *Sclerotinia* infection.

Impacts of solarization on the soil microbiome in Pacific Northwest tree seedling nurseries

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Soil solarization is a pre-planting soil treatment that employs solar radiation to heat soil under a transparent plastic film to achieve temperatures detrimental to certain soilborne pathogens and weed seeds. Little is known about its effects on soil microbial communities. Replicated field trials with solarized and non-solarized plots were conducted at three Pacific Northwest tree seedling nurseries in July-August, 2016. At each site, nylon mesh sachets containing homogenized composite soil samples from that site were buried at 5 and 15 cm depths. Soil temperatures were monitored, and sachets were retrieved after six weeks of treatment. DNA was extracted from soil and PCR was performed using 16S (bacterial) and ITS1 (fungi & oomycetes) primers. Amplicons were then sequenced using the Illumina Miseq platform. Sequence data were compared to 16S and ITS reference databases for microbial identification and community analysis. Compared to the non-solarized treatment, solarization resulted in reduced alpha-diversity and species richness at the 5 cm depth, but not the 15 cm depth, likely reflecting soil temperature effects. We also identified bacterial, fungal and oomycete taxa significantly impacted by soil solarization. Our study improves the understanding of soil solarization impacts on soilborne pathogens and other soil microbes that could directly or indirectly influence plant growth.

Prediction of Verticillium wilt in mint fields using *Verticillium dahliae* DNA, *Pratylenchus penetrans*, mint cultivar, and crop age

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Verticillium wilt reduces yields in mint fields. Management of wilt is limited because *Verticillium dahliae* (*Vd*) survives in soil for over a decade, synergistically interacts with the root-lesion nematode *Pratylenchus penetrans* (*Pp*), infects a wide range of crops, and resistance is not available in most mint cultivars. Risk assessment tools are required to inform management decisions concerning field selection and soil fumigation before planting. The objective of this research was to develop a model to predict the occurrence of wilt in mint fields. Soil samples were collected from 96 study sites in 30 mint fields in Oregon and Washington. *Vd* and *Pp* were quantified with quantitative real-time PCR or counted, respectively, and wilted stems were subsequently quantified in fields. Parametric and kernel-based non-parametric models were fit to the data, compared, and cross-validated. Although the parametric model was not misspecified ($J_n = 0.11$, $P = 0.07$), the non-parametric model had a slightly greater multiple R^2 (0.58 vs 0.53) and a lower predicted square error (3.5 vs. 25.7) than the parametric model. Both models identified estimates of *Vd* DNA, counts of *Pp*, mint cultivar, and crop age as predictors of wilt in mint fields. These models (i) revealed the quantitative aspects of the four-way interaction between wilt, two pathogens, host cultivar, and host age and (ii) provide a foundation on which predictive forecasting tools can be developed for mint growers.

Physiological effects of sub-lethal doses of post-plant nematicides on *Meloidogyne incognita*

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Meloidogyne species are one of the most important groups of plant-parasitic nematodes globally due to their ability to damage most cultivated plants. Although widespread and economically important, there are few control measures to combat these nematodes. New nematicides were discovered which have the potential to be widely utilized for nematode management. For proper stewardship and long-term efficacy of these nematicides, more information is required regarding physiological impacts of these compounds on nematodes. In this study, post-plant nematicides: Vydate, Salibro, Nimitz, and Velum, were used to understand how sub-lethal doses affect fecundity and mobility of *Meloidogyne incognita* second stage juveniles (J2). Using a microwell assay, dose-response curves for each nematicide were established for *M. incognita* J2. Velum was the most toxic nematicide, with effective doses up to 230 times lower than that of other nematicides. The 24-hour dose of each nematicide that resulted in the mortality of 50% of the population (LD₅₀) was then used in both mobility and infectivity assays. *M. incognita* J2 exposed to a LD₅₀ dose of Salibro and Nimitz had reduced viability of mobile J2. Whereas, J2 exposed to Velum and Vydate were not different from the control. *M. incognita* J2 exposed to a LD₅₀ doses of Salibro, Vydate, and Nimitz were unable to reproduce on tomato, unlike *M. incognita* J2 exposed to Velum, which had reproduction capability similar to that of the control.

AmplifyRP® - a versatile diagnostic tool for rapid detection of plant pathogens

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Many plant pathogens can cause serious diseases in plants and significant economic losses. Thus, it is very important to have early detection and effective control of plant pathogens to prevent from wide spreading. AmplifyRP®, a registered trademark of Agdia Inc.,

utilizes a leading isothermal amplification technology called recombinase polymerase amplification (RPA) and has recently become a versatile diagnostic tool for rapid and accurate detection of nucleic acids in all types of plant pathogens such as fungi, bacteria, viruses or viroids. In this presentation, we will review how AmplifyRP® works and what kinds of test formats and products are available at present. In brief, all the specific recombination and target amplification in AmplifyRP® assays occur at a constant temperature of 39°C in a single PCR tube and a test can be completed from sample to result within 30 minutes or less. The test result can be displayed either in real-time with a portable fluorometer such as AmpliFire or on a lateral flow strip contained within a contamination-proof amplicon detection chamber or both. AmplifyRP® assays can use simple crude plant extracts as amplification templates and yet are as sensitive as PCR. AmplifyRP® is applicable both in the laboratory and in the field.

Determining communities within the pea root rot complex and their associations among soil edaphic properties and disease incidence

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Field pea production is greatly impacted by multiple root rotting fungal and oomycete pathogens referred to as the pea root rot complex (PRRC). The objectives of this research were to characterize soil-borne pathogen communities (groups) within the PRRC; and establish associations and develop prediction models among soil edaphic properties, occurrence of each PRRC community and disease incidence. Hierarchical clustering, indicator species analysis, and multi-response permutation procedures identified five communities, characterized by (1) *Fusarium acuminatum*, (2) *Fusarium solani*, (3) *Aphanomyces euteiches*, (4) *Fusarium sporotrichioides*, and (5) *Fusarium redolens*. Logistic regression was not performed for communities 2 and 5 due to insufficient data to make any significant or meaningful associations. Results from logistic regression indicated that communities 1 ($p < 0.01$) and 4 ($p < 0.01$) were associated with clay content. Soil pH ($p = 0.01$), Fe^{2+} ($p < 0.01$) and K^+ ($p = 0.02$) were associated with the occurrence of community 3. Soil pH ($p = 0.01$) and Fe^{2+} ($p = 0.03$) were associated with community 4. Disease incidence increased in the presence of either community 1 or 3. Increasing K^+ levels decreased the probability of diseased plants being present. Information generated from this research will contribute to the development of PRRC management strategies by providing a soil-borne pathogen community prediction tool.

Searching for resistance to soilborne pathogens in cultivated strawberries and the *Fragaria* supercore

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Soilborne pathogens are a challenge for strawberry (*Fragaria ×ananassa*) growers. Pre-planting soil fumigation is used to control *Verticillium dahliae* (*Vd*), *Macrophomina phaseolina* (*Mp*), and *Fusarium oxysporum* f.sp. *fragariae* (*Fof*). Fumigation is detrimental to microbial communities. Genetic resistance can be an economical, environmentally friendly option for disease management. To identify sources of disease resistance, 21 *F. ×ananassa* accessions and 32 individuals from the *Fragaria* supercore, composed of *F. chiloensis* and *F. virginiana* accessions, were evaluated. Six plants of each accession were inoculated via separate root dips prior to planting. Percent mortality was recorded after 10 weeks and a mortality less than 33.3% was considered resistant. Fifteen *F. ×ananassa* accessions were resistant to *Vd*, eight were resistant to *Mp*, and 12 were resistant to *Fof*. MI 10-24-52 was the only *F. ×ananassa* accession resistant to all three pathogens. From the supercore, 13 of 32 accessions evaluated were resistant to *Fof*, 18 of 23 were resistant to *Vd*, and 13 of 16 were resistant to *Mp*. Out of 16 accessions evaluated with each pathogen, eight were resistant to all three. Six *F. ×ananassa* bi-parental populations have been developed to identify genes involved in resistance to these pathogens. Future work will focus on identifying resistance genes, developing tools for DNA-informed breeding, and introgressing resistance from the supercore into *F. ×ananassa*.