Abstracts of Special Session Presentations

Biology of Plant Pathogens

Coevolution of Fungi and Plants

Fungal/plant interactions during the early terrestrialization of the land. T. N. TAYLOR. University of Kansas. Phytopathology 94:S119. Publication no. P-2004-0001-SSA.

The first putative fungi occur in Silurian rocks and consist of hyphae and possible phialides. Ordovician resting spores have recently been reported, but the age and provenance of these is equivocal. These geologically early fungal remains are dispersed in the rock record and recovered from macerations and thus it is impossible to evaluate their interactions with other organisms. To date the first demonstration of fungi biologically associated with other organisms occurs in the Early Devonian Rhynie chert from Scotland. In this 400 million-year-old fresh water, hot springs environment are several fungal groups (Chytridiomycota, Ascomycota, Zygomycota). The exquisite preservation of these fossils makes it possible to document a variety of interactions between fungi and early land plants comparable with those in modern ecosystems. These organisms show evidence of fungal invasion and illustrate responses by the hosts. Fungi from the Rhynie chert indicate the rapid evolution of thallus type and nutritional mode, as well as complex life history biology and host interactions. Using the Early Devonian as a calibration point in fungal evolution, it becomes obvious that many of the organisms considered today as fungi have remained relatively unchanged since that time. As a major component of the microbial world during the early terrestrialization of the earth, understanding early fungi is especially important in helping to frame hypotheses to investigate how these organisms contributed to the evolution and stability of the terrestrial ecosystem.

Evolutionary dynamics of the interactions between plants and arbuscular mycorrhizal fungi. J. BEVER. Indiana University. Phytopathology 94:S119. Publication no. P-2004-0002-SSA.

Many plant species take up soil minerals such as phosphorus through mutualistic interactions with arbuscular mycorrhizal fungi. While this mutualism is of obvious ecological importance, our knowledge of the dynamics within this mutualism is limited. In this talk, I will develop and test two general frameworks for understanding evolution of this interaction. I first use general coevolutionary models to identify that either positive or negative frequency dependent dynamics could occur within the mycorrhizal mutualism. Positive frequency dependent dynamics are generated by symmetric fitness relationships and predict a reinforcement and strengthening of the mutualism, while negative frequency dependent dynamics result from highly asymmetric fitness relationships and creates a more active dynamic that fails to maximize the benefit from the interaction. In testing these dynamics, I found evidence of negative frequency dependence which would contribute to the maintenance of diversity within the system, but also leads to a decline of mutualistic benefit over time. The degradation of the plant-mycorrhizal mutualism would also be predicted as an evolutionary consequence of any costs the fungus endures in delivering phosphorus to its host. I develop a second framework to understand what maintains the mutualism in the face of these evolutionary pressures. Specifically, I predict that the mutualism will be maintained provided that plants can allocate preferentially to the most beneficial fungus. I finally present experimental results that support this hypothesis.

Host specialization of endophytes and coevolution. C. SCHARDL. University of Kentucky. Phytopathology 94:S119. Publication no. P-2004-0003-SSA.

Symbioses of grasses with *Epichloë* endophytes represent a continuum from mutualistic to antagonistic associations. In most cases, each phylogenetic species of endophyte is host specific, confined to one grass species, genus, or group of closely related genera. Comparative phylogenetic analysis of the grass subfamily Poöideae with *epichloë* endophytes suggests that there may have been a history of coevolution (co-cladogenesis) over tens of millions of years. A notable exception, however, is the broad host-range and genetically diverse species, *Epichloë typhina*. A population genetic study of *E. typhina* nevertheless indicates that host-specialized populations are genetically isolated from each other, thus constituting cryptic species. Interestingly, some such populations appear to be polyphyletic. A common assumption of phylogenetic species recognition is that speciation is followed by lineage sorting, eventually leading to monophyletic species. Apparently, within the *E. typhina* cryptic species complex, lineage sorting has not necessarily resulted in monophyly even after several million years of divergence. I will discuss whether these unexpected evolutionary patterns may relate to coevolution and co-adaptation of endophytes and their hosts.

The human impact on plant disease, it's not all bad. G. MAY. University of Minnesota. Phytopathology 94:S119. Publication no. P-2004-0004-SSA.

The landscape and the ecology of the Americas has been profoundly affected by the domestication of maize and expansion of maize populations over the last 8,000 years. Such dramatic changes under human management are often associated with accelerated evolution of virulence in pathogens countered by resistance in plant hosts. Despite development of strong resistance in the early 1900's, resistance to corn smut has been 'durable'. In our research, we ask whether gene-for-gene warfare is an inevitable consequence of crop breeding and management. We present evidence for the genetic basis of the durable but complex resistance traits in maize to *Ustilago maydis* (corn smut) and for the evolutionary response of the pathogen to host domestication and population expansion.

Phylogeographic studies on tree pathogens reveal differing patterns of speciation and host pathogen co-evolution. B. D. WINGFIELD, M. P. A. Coetzee, and M. J. Wingfield. University of Pretoria. Phytopathology 94:S119. Publication no. P-2004-0005-SSA.

Ready access to DNA sequence data has made it possible to explore patterns of speciation and host pathogen co-evolutions at levels that have previously not been possible. Contemporary studies on various plant pathogenic ascomycetous fungi suggest relatively recent speciation events and consequent adaptation to new hosts and environments. Our studies, for example on *Cryphonectria cubensis*, the causal agent of a serious canker disease of Eucalyptus supports this view. Our studies on *Armillaria* spp. that are important root pathogens of forest trees in many parts of the world have yielded contrary results. Thus, emergent phylogenetic trees separate the species into two strongly supported groups representing the Holarctic and the non-Holarctic (African, Australian, Indo-pacific and South American) Floral Kingdoms. The estimated time of divergence of these groups suggests that *Armillaria* spp. from non-Holarctic are considerably older than those from the Holarctic. Furthermore, the non-Holarctic species appear to have originated in Gondwana. Speciation in these strongly clonal root pathogens appears to be strongly linked to vicariance. The implication of this finding is that generic and species boundaries in some fungal species will be quite different to those

in others. This will impact both on the characters that we use to define these boundaries as well as on our understanding of host pathogen co-evolution.

Geological time, evolutionary rates and the history of fungi. J. W. TAYLOR. University of California, Berkeley. Phytopathology 94:S120. Publication no. P-2004-0006-SSA.

Phylogenetic trees that emphasize the "big-picture" of fungal evolution have been made from ribosomal DNA sequences and from amino acid alignments of protein coding genes, and both types have been fitted to a geological time scale. The tree topologies are similar, but there is controversy over the timing of divergences based upon which date is taken for the split between animals and fungi, i.e., 900 to 1.6 billion years ago. Often, the earliest date for the appearance of a fungal lineage is much older than the earliest available fossil. This discrepancy can be explained by the difficulty of finding fungal fossils, the difficulty in recognizing members of fungal lineages soon after divergence, and bias in phylogenetic tree construction. It is hard to date recent events in fungal evolution with the "big picture" approach because rDNA variation or amino acid variation is small over short periods. For recent events, DNA sequence of protein coding genes is useful if nucleotide substitution rates can be determined for these genes. One approach is to use the "big picture" tree to calibrate protein trees that emphasize smaller clades of interesting fungi. This approach has been demonstrated for Eurotiomycetes (Plectomycetes) and has shown that estimates of nucleotide substitution rate are strongly influenced by the use of maximum likelihood or distance methods, by whether or not variation in substitution rate on different branches is permitted, and by the date of the animal-fungal divergence. Using the estimated rate of nucleotide substitution for protein coding genes, possible recent interactions between humans and their fungal pathogens have been examined.

Current Insights Into the Genetics, Toxicology, and Plant Pathology of *Fusarium verticillioides*

Perspectives on the history and taxonomy of *Fusarium verticillioides* (teleomorph, *Gibberella moniliformis*). A. E. DESJARDINS. NCAUR/ARS/USDA, Peoria, IL 61604. Phytopathology 94:S120. Publication no. P-2004-0007-SSA.

In Nebraska in 1904, Sheldon first described *Fusarium moniliforme* from moldy maize in feed associated with animal diseases. This anamorph name was used widely by plant pathologists throughout the twentieth century and, in 1924, Wineland described the teleomorph as *Gibberella moniliformis*. Sheldon based the species epithet on the Latin *monilis*, meaning necklace, in reference to the distinctive long chains of microconidia. In Italy in 1881, Saccardo described *Oospora verticilloides*, a microconidial-chain producing fungus, from maize associated with the human disease pellagra. Scientists were soon aware that these two maize pathogens were identical, and in 1976 Nirenberg combined *F. verticillioides/G. moniliformis*, the presently accepted nomenclature. A combination of morphological, biological, and phylogenetic species approaches is resolving *F. verticillioides* and other agriculturally important species of the *G. fujikuroi* species complex. *F. verticillioides* remains the most important mycotoxigenic species of this complex due to its production of carcinogenic fumonisins and its widespread occurrence on maize, an American crop plant that now feeds human populations throughout the world.

The fumonisin risk assessment and emerging issues. R. T. RILEY and K. A. Voss. USDA-ARS, Athens, GA. Phytopathology 94:S120. Publication no. P-2004-0008-SSA.

Fumonisins pose a tortuous problem for risk assessment. Unlike most mycotoxins, fumonisins of the B series (FB) are water soluble and unlike many other carcinogens are not metabolized nor react directly with DNA. Their toxicity is species-, sex-, and strain-dependent. FB1 was the first naturally occurring compound shown to inhibit ceramide biosynthesis. Ceramide is an intermediate in, what was in 1990, an obscure lipid pathway. One of the first effects of FB1 is increased apoptosis; a process normally considered to reduce cancer risk. FB is rapidly excreted and poorly absorbed; yet evidence of exposure can persist after FB is removed from the diet. *Fusarium verticillioides* was once described as a "Mycotoxicological Miasma". It should have been no surprise that structurally FBs are sphingolipid-like and sphingolipids are the "Sphinx" of lipids. This presentation will include an historical account of the FB risk analysis from the recognition that moldy maize caused farm animal diseases until the first cancer evaluation of FB1 was completed in 2002. Nonetheless, although the USFDA has issued official guidance to industry, the fumonisin risk assessment is far from complete. For example, some studies suggest that FBs are not teratogenic and that fetal toxicity, when present is secondary to maternal toxicity. More recent data suggests, however, that neural tube defects can be induced in one mouse strain and the incidence is closely linked to altered folate receptor function. Thus, future issues could surface and they must be resolved if science-based risk assessment is to be the basis for future re-evaluation of regulatory guidelines or tolerances.

Elucidation of the biosynthetic pathway by deletion analysis. R. A. E. BUTCHKO, R. H. Proctor, and R. D. Plattner. NCAUR/ARS/USDA, Peoria, IL 61604. Phytopathology 94:S120. Publication no. P-2004-0009-SSA.

Fumonisins are polyketide mycotoxins produced by the maize pathogen *Fusarium verticillioides*. A fumonisin biosynthetic gene cluster, consisting of 15 co-regulated genes, has been described in *F. verticillioides*. Predicted amino acid sequences of these genes suggest that most are involved in fumonisin biosynthesis. To determine the role of these genes, we deleted or disrupted each gene and examined the resulting effect on fumonisin production. The deletion/disruption mutants fell into four phenotypic classes: 1) complete loss of fumonisin production, 2) loss of certain fumonisins, 3) accumulation of novel fumonisin homologues, and 4) normal fumonisin production. These analyses along with feeding studies with the novel fumonisin homologues have allowed us to assign genes/enzymes to individual fumonisin biosynthetic reactions and as a result to propose a fumonisin biosynthetic pathway.

Genomics approach for solving a mycotoxin problem in maize. R. H. PROCTOR (1), D. W. Brown (1), R. A. E. Butchko (1), R. D. Plattner (1), and D. Kendra (2). (1) USDA ARS NCAUR, Peoria, IL; (2) TIGR, Rockville, MD. Phytopathology 94:S120. Publication no. P-2004-0010-SSA.

Fumonisins are polyketide-derived mycotoxins produced by the maize pathogen *Fusarium verticillioides* and cause several fatal animal diseases including cancer in laboratory rodents. Eliminating fumonisins in maize has been a major objective of fumonisin-related research since the identification of these mycotoxins in 1988. To achieve these objectives, researchers are trying to elucidate the genetic and biochemical pathways that regulate fumonisin biosynthesis and to identify factors that allow *F. verticillioides* to infect maize and cause disease. As part of these approaches, we have created a *F. verticillioides* Expressed Sequence Tag (EST) database. The ESTs were generated from nine cDNA libraries, each constructed from a different growth condition. Conditions included a liquid culture that promoted fumonisin production and a culture that contained maize tissue or extracts. To date over 92,000 cDNA clones have been sequenced. Bioinformatic analyses have revealed that these sequences correspond to over 9,500 *F. verticillioides* genes, including all fifteen genes in the fumonisin biosynthetic gene (*FUM*) cluster. The *FUM* ESTs were recovered only from cDNA libraries generated from fumonisin-promoting cultures. These results indicate that the EST database is rich in sequences of differentially expressed genes and that comparisons of ESTs derived from different cDNA libraries may facilitate the identification of fumonisin regulatory and maize-induced genes.

Effect of microenvironment on fumonisin biosynthesis. C. P. WOLOSHUK. Purdue University, West Lafayette, IN. Phytopathology 94:S120. Publication no. P-2004-0011-SSA.

As *Fusarium verticillioides* colonizes a maize kernel, initiation of fumonisin B1 (FB1) biosynthesis is influenced by kernel composition as related to tissue type, genotype and developmental stage. We have found that FB1 production during colonization is greater in the endosperm tissue than in germ tissue, greater in dent hybrids than in lines homozygous for the shrunken-2 allele, and greater in the dent stage of development than in the blister, milk and dough stages. We also have studied a number of regulatory genes in *F. verticillioides* that impact FB1 biosynthesis, such as Pac1 (pH regulator), Fcc1 (C-type cyclin), Fck1 (cyclin-dependent kinase), and Zfr1 (transcription factor), as well as expression profiles of *F. verticillioides* genes by microarray techniques. Not surprisingly, the molecular mechanisms that regulate FB1 biosynthesis appear complex. However, the evidence suggests that fungal metabolism of maize kernel components, especially amino acids and starch, plays an important role in the regulation of FB1 biosynthesis.

Identification and potential use of genetic resistance to control fumonisin accumulation in corn grain. D. G. WHITE (1), C. E. Kleinschmidt (1), M. J. Clements (2), and J. K. Pataky (1). (1) University of Illinois, Department of Crop Sciences, Urbana, IL; (2) USDA-ARS Corn Host Plant Resistance Research Unit, Mississippi State, MS. Phytopathology 94:S120. Publication no. P-2004-0012-SSA.

Commercial corn hybrids grown in environments that favor Fusarium ear rot and fumonisin production often produce grain with unacceptably high

concentration of fumonisin. Resistance must be identified from novel genetic sources and incorporated into commercial germplasm. F1 hybrids were developed with a diverse collection of over 1,500 inbreds and the susceptible, commercially-used inbred, FR1064. The F1 hybrids were evaluated in inoculated trials in Illinois and in naturally infected trials in North Carolina. Several of the F1 hybrids were highly resistant to fumonisin accumulation in grain. Resistant F1 hybrids were developed with inbreds representing a variety of endosperm types and maturities. Inheritance of resistance from these inbreds is being evaluated with generation means analyses. Quantitative trait loci associated with resistance from these inbreds are being identified with molecular markers and populations composed of 250 or more families of the backcross susceptible self generation. Our research strongly suggests that levels of resistance necessary to minimize fumonisin accumulation in grain in most environments can be incorporated into commercial germplasm, possibly through marker-assisted selection.

Functional Genomics Meets Bacterial Diseases

***Pseudomonas syringae*: A cosmopolitan pathogen with varied interactions with plants?** S. LINDOW. University of California, Berkeley. Phytopathology 94:S121. Publication no. P-2004-0013-SSA.

Strains of *P. syringae* can infect a large number of plant species and produce symptoms as diverse as leaf spots, cankers, blast, blisters, and galls. Since most investigations of virulence factors have emphasized interactions of the pathogen with leaf tissue, little is known of traits that might contribute to other types of infections. Many different sources of inoculum are possible for these diseases, yet little is known of traits that contribute to growth and survival of *P. syringae* in seeds, the spermosphere, phyllosphere or as an endophyte. It seems likely that many genes required for associations of the pathogen with potential host plants as well as in reservoir sites, in addition for that of the infection process itself, will exhibit a habitat-specific pattern of expression. Studies that address in planta gene expression of *P. syringae* will be summarized. The comparative epidemiology of diseases caused by different pathovars of *P. syringae* will be explored from the perspective of illustrating differences in the various phases of the infection process and to reveal research needs and opportunities to better understand this diverse pathogen.

***Pseudomonas syringae* pathogenicity explored from the perspective of type III secretion systems and comparative genomics.** A. COLLMER (1), J. R. Alfano (2), C. R. Buell (3), S. Cartinhour (4), A. K. Chatterjee (5), G. B. Martin (6), D. J. Schneider (4), and X. Tang (7). (1) Cornell University, Ithaca, NY; (2) The Institute for Genomic Research, Rockville, MD; (3) University of Nebraska, Lincoln, NE; (4) USDA-ARS, Ithaca, NY; (5) University of Missouri, Columbia, MO; (6) Boyce Thompson Institute for Plant Research, Ithaca, NY; (7) Kansas State University, Manhattan, KS. Phytopathology 94:S121. Publication no. P-2004-0014-SSA.

The Hrp type III secretion system (TTSS) of *P. syringae* plays a key role in the bacterium's host-specific pathogenicity. The TTSS injects virulence effector proteins into plant cells. More than 50 candidate TTSS effector genes have been identified in the complete genome sequence of *P. syringae* pv. *tomato* DC3000, a pathogen of tomato and Arabidopsis, based on the presence of characteristic promoter and N-proximal amino-acid patterns (Buell et al. 2003. Proc. Natl. Acad. Sci. USA 100:10181-10186). A draft sequence of the genome of *P. syringae* pv. *phaseolicola* 1448A has been obtained by The Institute for Genomic Research and is available through http://pseudomonas-syringae.org. The genome is currently being closed and then will be annotated and deposited into GenBank and the TIGR Comprehensive Microbial Resource. Refined experimental and computational tools for identifying TTSS effector genes are being applied to the 1448A genome. Comparison of the effector inventories, genomic islands, Hrp regulons, and mobile genetic elements of DC3000 and 1448A will provide a foundation for addressing many unanswered questions regarding the mechanisms and evolution of *P. syringae* pathogenicity.

***Pseudomonas syringae* DC3000: Old pathogen, new name?** C. BENDER. Oklahoma State University. Phytopathology 94:S121. Publication no. P-2004-0015-SSA.

P. syringae pv. *tomato* DC3000 and *P. s.* pv. *maculicola* ES4326 have been used as 'model' organisms in many studies largely because they are pathogens of Arabidopsis. However, both strains are pathogenic on tomato and edible *Brassica* spp. and can elicit very different symptoms on these hosts. The host range of both pathovars is similar, which raises interesting questions about the naming of pathovars and whether this issue should be revisited. *P. s. tomato* and *P. s. maculicola* are seedborne pathogens, an aspect of their biology and epidemiology that has applicability to high-throughput assays for pathogenicity and virulence. There is a wealth of literature pertaining to the taxonomy, epidemiology, and biology of pvs. *tomato* and *maculicola*, and this will be reviewed in the context of genomics and research initiatives for the future.

The *Ralstonia solanacearum* species complex: Genetic diversity and physiology of the pathogen and ecology of bacterial wilt. A. C. HAYWARD (1) and M. Fegan (1,2). (1) Department of Microbiology and Parasitology, The University of Queensland, Australia; (2) Cooperative Research Centre for Tropical Plant Protection. Phytopathology 94:S121. Publication no. P-2004-0016-SSA.

The *Ralstonia solanacerum* species complex, including *R. syzygii* and the cause of blood bacterial wilt of banana, is a subgroup within the beta-proteobacteria based on sequence differences in the 16S rDNA genes. Sequence differences in pathogenicity related genes and other conserved regions of the genome have shown that there are four phylogenetic lineages (phylotypes). Most of the detailed studies on host-parasite interactions have been made with the strains K60 and AW1 of phylotype II and with GMI1000 of phylotype I. Bacterial wilt affects representatives of more than 50 plant families and is primarily a disease of the tropics and subtropics which, in the case of certain strains of the pathogen and particular hosts, has spread into warm temperate and cool temperate regions on latently infected planting material. Strains of the pathogen differ in host specialisation and environmental adaptation. For example,bacterial wilt of tomato requires a high diurnal temperature range for symptom development; by contrast on potato latent infections are prone to establish on progeny tubers at lower temperature regimes without wilt symptoms on stems and leaves. Modes of dissemination and transmission are diverse. The disease is spread by soil and water, by root-to-root contact and by mechanical means; there is little information on survival as an epiphyte or of transmission on true seed. Bugtok disease of cooking banana (ABB/BBB genotype) in the Philippines caused by *R. solanacearum* race 2 is a fruit rot spread by insects to the inflorescence and wilt symptoms are absent; the same pathogen causes a wilt (Moko disease) on Cavendish dessert banana. The variable physiology and ecology of members of the complex and key areas for future research will be highlighted.

Pathogenicity determinants and genomics of *Ralstonia solanacearum*. S. GENIN, A. Angot, S. Cunnac, A. Occhialini, N. Peeters, and C. Boucher. LIPM, INRA-CNRS, BP27, 31326 Castanet-Tolosan, France. Phytopathology 94:S121. Publication no. P-2004-0017-SSA.

As many other important plant and animal pathogens, *R. solanacearum* uses a Type III secretion system (TTSS) to inject virulence effector proteins into host cells. From the complete genome sequence of *R. solanacearum* strain GMI1000 (race1, wide host range), we have used a combination of computational and experimental approaches as a first step for establishing an inventory of TTSS-effector proteins. We have identified > 40 TTSS-effectors and have started their functional analysis. Three effector genes, popP1, popP2 and avrA, were found to encode avirulence factors against Petunia, Arabidopsis and tobacco, respectively. In particular, popP2 is the matching avirulence gene of the TIR-NBS-LRR-WRKY resistance gene RRS1-R conferring resistance to several strains of the pathogen. Finally, GMI1000 whole genome microarrays have been generated and were used to identify the repertoire of regulatory targets of HrpB, a key regulator of pathogenicity. Transcriptomic analyses using such regulatory mutants should identify novel groups of target genes contributing to virulence.

Extracellular space: The final frontier for *Ralstonia solanacearum* proteins required for colonization. T. P. DENNY, H. Liu, J. J. Wolff, I. J. Amster, and M. A. Schell. The University of Georgia, Athens, GA. Phytopathology 94:S121. Publication no. P-2004-0018-SSA.

The majority of extracellular proteins (EXPs) secreted by R. solanacearum probably transit its *sec*-dependent type II pathway rather than the *sec*-independent type I or III pathways. The type II pathway also is much more important than the type III pathway for invasion and systemic colonization of tomato plants by *R. solanacearum*. Relatively little is known about EXPs that transit the type II pathway in Gram-negative bacteria, in part because they cannot be predicted based on analyses of DNA or protein sequences available in a genomic database. Therefore, we are using genetic and proteomic approaches to study the complete set of EXPs in culture supernatants and to identify the subset of proteins necessary for colonization. A catalog of *R. solanacearum* EXPs is being created using a combination of three strategies:

(i) standard 2-dimensional gels and MALDI-TOF mass spectrometry, (ii) 'shotgun' peptide analysis using liquid chromatography and Fourier transform ion cyclotron resonance mass spectrometry, and (iii) high throughput epitope tagging and immunological detection of EXPs. To demonstrate the involvement of a cadre of EXPs in colonization, we optimized the pyramiding of unmarked gene deletions in *R. solanacearum*. Unlike a type II pathway mutant, a mutant of GMI1000 unable to produce any of the six known plant cell wall-degrading enzymes is colonization competent.

Genome-Based Studies of Fungal–Plant Pathosystems

Functional genome analysis of *Magnaporthe grisea*. R. A. DEAN, T. Mitchell, D. Brown, N. Donofrio, and Y. Oh. North Carolina State University Fungal Genomics Laboratory, Raleigh, NC. Phytopathology 94:S122. Publication no. P-2004-0019-SSA.

Rice blast, caused by the fungus *Magnaporthe grisea*, is historically the most devastating disease of rice and a threat to global food supplies. In collaboration with six other researchers, we are working to characterize putatively secreted proteins from the fungus to identify those that interact with the plant, generate >50,000 random integration fungal mutants to identify genes required for reproduction and pathogenicity, sequence 35,000 ESTs from infected and uninfected rice tissues, analyze six different rice Long SAGE libraries, generate and using over 400 interactions microarray chips containing rice blast as well as rice genes, and perform targeted gene deletions of genes in the fungus we predict to be required for pathogenicity. All data generated by this project is collected in a centralize database called MGOS, and is presented to the public through a web interface that will allow researchers to query the data. The current status of the project will be presented with a concentration on results from gene expression analysis.

Genomics of the wheat and barley pathogen, *Fusarium graminearum*. H. C. KISTLER (1), F. Trail (2), B. Birren (3), L. Ma (3), J. Galagan (3), L. R. Gale (1), K. O'Donnell (4), K. Seong (5), and J.-R. Xu (5). (1) USDA ARS Cereal Disease Laboratory, St. Paul, MN; (2) Michigan State Universty, East Lansing, MI; (3) Broad Institute, MIT, Cambridge, MA; (4) Microbial Genomics Research Unit, USDA ARS, Peoria, IL; (5) Purdue University, West Lafayette, IN. Phytopathology 94:S122. Publication no. P-2004-0020-SSA.

Head blight or scab caused by *Fusarium graminearum* is a disease of wheat and barley that occurs worldwide and emerges as the plant disease with great impact on U.S. agriculture and society. Infested cereals are often contaminated with trichothecene and estrogenic mycotoxins. To better understand fungal pathogenesis and development in this important pathogen, we have generated over 10,000 ESTs from three cDNA libraries and a draft sequence assembly of the *F. graminearum* genome. Using organism-specific parameters for gene prediction, 11,640 protein-coding genes have been identified. The entire assembled 36 Mb genome sequence consists of just 28 supercontigs (496 contigs > 2 kb). A genetic map, consisting of 131 SNPs, and 27 microsatellites, has been constructed that anchors 99.9% of the sequence assembly. Random insertional mutagenesis and targeted gene disruption approaches have been applied to identify genes important for plant infection and fungal growth. Details of automated and manual annotation, the whole-genome microarray, and coordination of functional analyses will be discussed. The *F. graminearum* sequencing and microarray projects were funded by the USDA/NSF Microbial Genome Sequencing Program and UDSA Integrated Program.

Genome sequencing and gene expression in *Aspergillus flavus*. G. A. PAYNE and M. S. Price. North Carolina State University, Raleigh, NC. Phytopathology 94:S122. Publication no. P-2004-0021-SSA.

Aspergillus flavus is an opportunist pathogen of plants and animals. While not considered an aggressive pathogen, it can parasitize the developing seeds of diverse plant species, including corn, peanut, cotton, and tree nuts. A consequence of this growth is the production of the carcinogen, aflatoxin. To better understand the growth and metabolism of this fungus, a targeted array of 768 elements whose transcription is induced during aflatoxin biosynthesis is being used to study gene expression. Four culture conditions comparing conducive and nonconducive culture pH and temperature, and carbon and nitrogen sources were examined for differential gene expression. The conducive conditions and numbers of upregulated genes were: sucrose (270); nitrate (145); pH 4.5 (242); 28 C (94). Only 9 genes were upregulated and 1 gene was downregulated in all four conditions. When the data for all conditions were analyzed by hierarchical clustering, the expression profile of all the structural pathway genes clustered together except *nor1*. The regulatory genes *aflJ* and *aflR* did not cluster with each other or with any gene known to be involved in aflatoxin biosynthesis. Several genes with no known function also clustered with the aflatoxin biosynthetic genes and these genes are being examined for their role in aflatoxin biosynthesis.

The use of microarrays for the identification of virulence genes in *Ustilago maydis*. R. KAHMANN, H. Eichhorn, F. Lessing, and P. Mueller. Max Planck Institute for Terrestrial Microbiology, Marburg, Germany. Phytopathology 94:S122. Publication no. P-2004-0022-SSA.

Ustilago maydis is a dimorphic fungus that switches from a yeast-like haploid stage to a filamentous dikaryon after mating of compatible strains. In nature it is the dikaryon that is able to differentiate infection structures and cause disease on corn plants. In this system cAMP signaling as well as two MAP kinase modules regulate discrete steps during pathogenesis. To identify downstream targets of these signaling pathways we have applied molecular tools for functional genome analysis. In particular we have constructed strains where the individual pathways can be genetically activated. Changes in transcript levels were analyzed using Affymetrix chips representing about 6200 of the estimated 7000 *U. maydis* genes. Among the genes differentially regulated when the PKA was induced were 10 genes with putative functions in reductive and non-reductive iron uptake. Besides a related expression pattern these genes were clustered on three chromosomes and were all repressed by iron. *fer2*, encoding a high affinity ferric permease,was analyzed in detail. *fer3* mutants show a selective growth disadvantage on medium containing low amounts of Fe^{3+} iron and this can be complemented by addition of the two siderophores normally produced by *U. maydis*. *fer2* mutants are able to infect plants but disease symptoms are strongly attenuated. This demonstrates the importance of reductive iron uptake for virulence in *U. maydis*.

Proteomic identification and functional analyses of the *Magnaporthe grisea* secretome. S.-C. WU, V. S. K. Kolli, P. Albersheim, A. G. Darvill, and R. Orlando. Complex Carbohydrate Center, University of Georgia, Athens, GA 30602. Phytopathology 94:S122. Publication no. P-2004-0023-SSA.

Extracellular proteins (ECPs) participate in diverse and essential biological activities such as nutrition uptake, growth and cell-cell communication, in various cell types at different physiological states. Perhaps most importantly, some ECPs are known to play crucial roles in fungal pathogenicity and fungus-host interactions. High-throughput proteomics technologies such as 2-dimensional gel electrophoresis-mass spectrometry (2DE/MS) and multidimensional liquid chromatography-mass spectrometry (MDLC/MS) are being deployed to identify the total ECPs (designated as a "secretome") from *M. grisea* grown under various conditions. In addition to a large number of enzymes involved in plant cell wall catabolism and other known functions, many identified are novel proteins. Some of the ECPs may even be "host-specific". Preliminary results from the functional analyses of selected ECPs will be discussed in prospective of the current advances in proteomics research. [Supported by U.S. Department of Energy grants DE-FG05-93ER20221 and DE-FG02-93ER20097, the National Science Foundation grant NSF-9626835 and the National Institutes of Health grant P41RRR05351]

Comparison of Arabidopsis responses to *Alternaria brassicicola* and *Pseudomonas syringae* by expression profiling. S. VAN WEES (1), S. Goregaoker (1), and J. Glazebrook (1,2). (1) Torrey Mesa Research Institute, 3115 Merryfield Row, San Diego, CA 92121 (closed); (2) Department of Plant Biology, University of Minnesota, 1445 Gortner Avenue, St. Paul, MN 55108. Phytopathology 94:S122. Publication no. P-2004-0024-SSA.

Alternaria brassicicola strain MUCL20297 is a poor pathogen of Arabidopsis, failing to cause disease symptoms on any ecotype tested. Jasmonic acid signaling and the Arabidopsis phytoalexin, camalexin, are required for resistance, but salicylic acid signaling is not required. In contrast, resistance to the bacterial pathogen *Pseudomonas syringae* requires salicylate signaling but not jasmonic acid signaling or camalexin. Plant responses to these two pathogens were compared by expression profiling using an Affymetrix array representing one-third of the Arabidopsis genome. Gene expression changes occurred within 12 hours after *Alternaria* treatment. Approximately 50% of *Alternaria*-induced genes were also induced by *P. syringae*. Reverse genetics analysis of *Alternaria*-induced genes led to discovery of a cytochrome P450 monooxygenase required for camalexin synthesis and resistance to *Alternaria*.

Life Styles and Genomics of Fastidious and Gram-Positive Bacteria

A functional equivalent of a Type III secretion in Gram-positive bacteria. M. CAPARON. Dept. of Molecular Microbiology, Washington University School of Medicine, St. Louis, MO. Phytopathology 94:S123. Publication no. P-2004-0025-SSA.

Conventional wisdom asserts that because Gram-positive pathogens lack an outer membrane and lack Type III secretion, that the export of virulence proteins involved in manipulation of host cell behavior will be a simplified version of that found in Gram-negative pathogens. However, recent data from the human pathogen *Streptococcus pyogenes* does not support this hypothesis. These studies have shown that while this Gram-positive bacterium exclusively utilizes the General Secretory (Sec) pathway for protein export, that the Sec pathway coordinates the function of secretory and accessory folding factors using the ExPortal, a novel organelle that organizes the Sec machinery and accessory folding factors at a single unique microdomain of the cellular membrane. The ExPortal is a component of the Injectosome, a complex between the Sec pathway and a pore-forming cytolysin that functions to inject signal transduction proteins into host cells via a process known as Cytolysin-Mediated Translocation. Thus, Gram-positive pathogens have adapted the Sec pathway to function as a specialized secretion pathway for delivery of virulence proteins into host cells.

Evolution of plant pathogenicity in the genus *Streptomyces*. R. LORIA, J. A. Kers, M. V. Joshi, and E. G. Johnson. Department of Plant Pathology, Cornell University, Ithaca, NY. Phytopathology 94:S123. Publication no. P-2004-0026-SSA.

A polyphyletic group of plant pathogenic *Streptomyces* species cause scab diseases of tubers and other underground plant structures. Genetic analyses indicate that horizontal transfer of a large DNA fragment is the basis for emergence of new pathogenic species, including *S. turgidiscabies* and *S. acidiscabies*. The DNA fragment is self-transmissible, integrates into the chromosome of the recipient strain, and has the attributes of a pathogenicity island (PAI). The PAI contains the biosynthetic pathway for thaxtomin, a phytotoxin that inhibits cellulose synthesis in higher plants and is required for pathogenicity. Nec1, a secreted necrogenic protein, is encoded by a novel low G+C ORF on the PAI. The PAI also contains pathogenicity loci that are shared with other actinomycete plant pathogens. A tomatinase homolog is present on the *Streptomyces* PAI and in *Clavibacter michiganensis*: these proteins are plant pathogenicity factors in some fungi. The *fas* operon, first described from *Rhodococcus fascians*, is a recent addition to the *S. turgidiscabies* PAI.

The *Spiroplasma kunkelii* genome: Insights to a parasitic lifestyle in insects and plants. R. E. DAVIS (1), Y. Zhao (1), E. L. Dally (1), R. Jomantiene (1), S. Lin (2), B. Roe (2), and J. Shao (1). (1) USDA-Agricultural Research Service, Beltsville, MD; (2) University of Oklahoma, Norman, OK. Phytopathology 94:S123. Publication no. P-2004-0027-SSA.

We are sequencing the 1.6 Mbp genome of the helical, motile, cell wall-less prokaryote, *Spiroplasma kunkelii*, causal agent of corn stunt disease, to gain insights into its parasitism and pathogenicity in plants and insect vectors. Sequence data are available at http://www.genome.ou.edu/spiro.html. Features of the genome include genes found in the *Bacillus/Clostridium* group but absent in *Mycoplasma*; absence of cell wall biosynthesis genes found in the division/cell wall gene cluster of walled bacteria; numerous repeated sequences; and genes encoding adhesins, mobile elements including Spiroplasma virus and plasmids, and predicted macromolecule translocation systems: a Sec-dependent secretion pathway, a type IV secretion pathway, a twin-arginine pathway, a signal recognition particle-dependent pathway, and an ABC transporter-mediated pathway. The data provide clues to understanding evolutionary genome reduction approaching the minimal set of genes required for parasitism and pathogenicity in insect and plant hosts.

Phytoplasmas: From structural to functional genomics. X. Bai (1), V. Correa (1), J. Zhang (1), M. Goodin (3), S. Kamoun (2), and S. A. HOGENHOUT (1). (1) Department of Entomology; (2) Department of Plant Pathology, Ohio State University - OARDC, Wooster, OH; (3) Department of Plant Pathology, University of Kentucky, Lexington, KY. Phytopathology 94:S123. Publication no. P-2004-0028-SSA.

Phytoplasmas belong to the Class Mollicutes and are non-culturable intra- and extracellular pathogens of plants and insects. Various computer algorithms were employed to mine the near complete genome sequence of phytoplasma strain AY-WB for genes encoding potential virulence factors. So far, 56 candidate virulence proteins were selected for functional analysis. To determine whether these proteins affect plants, corresponding genes were cloned into the binary potato virus X expression vector, pGR106, and expressed in tomato and *Nicotiana benthamiana* via *Agrobacterium* mediated inoculation. This revealed 16 phytoplasma proteins that induced morphological aberrations in plants. Interestingly, five of the 16 proteins contain nuclear localization signals. Localization studies using yellow fluorescence protein (YFP) fusions showed that two of the five proteins target plant cell nuclei. Transcripts of genes of these two proteins were detected in plants during AY-WB infection.

The genome of the phytopathogenic bacterium *Clavibacter michiganensis* subsp. *michiganensis*. R. EICHENLAUB, K.-H. Gartemann, and A. Burger. University of Bielefeld, Bielefeld, Germany. Phytopathology 94:S123. Publication no. P-2004-0029-SSA.

The Gram-positive *Clavibacter michiganensis* subsp. *michiganensis* (Cmm) causes wilt and canker of tomato and has a circular genome of 3.3 Mbp (72.6% G+C) and two plasmids of 27.3 kbp (pCM1) and 72.5 kbp (pCM2). Both plasmids carry pathogenicity genes, *celA* (pCM1) encoding an endo-beta-1,4-glucanase and *pat-1* (pCM2) encoding a secreted serine protease. Plasmid curing results in an endophytic, symptom-less colonization of the host. A region of about 120 kb was identified on the chromosome containing seven ORFs homologous to *pat-1*, and several other genes for proteases, a tomatinase, and for polysaccharide hydrolysis and sugar uptake. The lower [G+C] of 65-68%, 1 kb direct repeats at the borders, and several plasmid/phage related genes indicate a "pathogenicity island". Field isolates defective in colonization of tomato had deletions of protease genes of this region. Furthermore, transposon mutants affecting putative protease genes were defective in colonization. Thus a chromosomal region carrying genes essential for interaction with the host plant has been identified.

Genome sequencing of *Clavibacter michiganensis* subsp. *sepedonicus*. C. A. ISHIMARU (1), D. L. Knudson (1), S. E. Brown (1), D. M. Francis (2), and J. Parkhill (3). (1) Colorado State University, Fort Collins, CO; (2) Ohio State University, Wooster, OH; (3) Sanger Institute, Wellcome Trust Genome Campus, Hinxton UK. Phytopathology 94:S123. Publication no. P-2004-0030-SSA.

The complete genome of *C. michiganensis* subsp. *sepedonicus* has been sequenced by a whole-genome shotgun strategy. This Gram-positive high GC coryneform bacterium causes bacterial ring rot of potato and is considered to be one of the most important bacterial pathogens of seed potatoes worldwide. The type strain, ATCC 33113, was chosen for sequencing because it is virulent and accessible to the scientific community. The GC content is 72.2-% and typical of the species. The genome is 3.5 Mb and contains a chromosome, circular plasmid (pCS1) and a linear plasmid (pCSL1). The current annotation indicates there are 65 copies of IS1121 and 25 copies of an IS1122-like element. Three additional repeat families, including four copies of IS30 have been detected. Several homologs of the pathogenicity (*pat-1*) locus from *Clavibacter michiganensis* subsp. *michiganensis* are also present. Progress on closure and annotation will be presented.

The interaction proteome of tomato and *Clavibacter michiganensis*. G. Coaker (1), W. Yang (1), B. Willard (2), M. Kinter (2), E. Stockinger (1), and D. FRANCIS (1). (1) The Ohio State University, 1680 Madison Ave., Wooster, OH 44691; (2) Cleveland Clinic Foundation, 9500 Euclid Ave., Cleveland, OH 44195. Phytopathology 94:S123. Publication no. P-2004-0031-SSA.

In tomato, resistance to *Clavibacter michiganensis* subsp. *michiganensis* (Cmm), is controlled by two QTL. Leaf tissue from lines containing *Rcm 2.0*, *Rcm 5.1*, and a susceptible control were compared using 2-DE through a time course to identify proteins that were differentially regulated in response to infection. The accumulation of specific proteins depended on genotype and time post-inoculation suggesting that the two QTL confer resistance through independent responses. Forty-seven tomato proteins were subjected to capillary column HPLC-tandem mass spectrometry. We were able to identify tomato genes or ESTs for 45 of 47 proteins. Proteins extracted from stem tissue offer an opportunity to identify bacterial proteins that are expressed in tomato during infection. The DNA sequence database for *Clavibacter michiganensis* subsp. *sepedonicus*, has provided a valuable tool to identify Cmm proteins. These bacterial proteins and the plant response to infection suggest possible mechanisms of virulence.

Microbial Forensics: Plant Pathogen Models

Plant pathogen forensics. W. T. COBB. Cobb Consulting Services. Phytopathology 94:S124. Publication no. P-2004-0032-SSA.

Dictionaries define the word *forensic* as "for the courts"; indicating the application of a particular subject to the law, as in forensic medicine, forensic accounting or, as in this case, some variation of forensic plant pathology. The application of our science to a courtroom setting is something that most of us have had no training for and probably little, if any, experience with. Judges, jurors and attorneys usually do not have strong science backgrounds, let alone any knowledge of even the existence of the science of plant pathology. Our contribution to the legal process is usually as either a "consulting" expert or as a consulting *and* testifying expert witness. Our job then is to make sure the science component of the proceeding is accurate, defensible and understandable to non-science trained individuals, be they a judge, a jury member or an attorney. Detection, diagnosis, verification and documentation of plant disease from the aspect of forensics will be discussed.

The forensics toolbox: Molecular clues, evolution and phylogeny. U. MELCHER. Oklahoma State University. Phytopathology 94:S124. Publication no. P-2004-0033-SSA.

Knowing how an outbreak of disease in one of our crops began is important for identifying the factors and/or persons responsible for the outbreak. Molecular tools can play a major role in uncovering the culprits. Forensic science begins when the causative pathogen species is already known. The task is to finger the one of many possible sources that is most likely to have caused the outbreak and to provide certainty estimates for that identification. A variety of methods can help. They include PCR fragment fingerprinting methods, microarray hybridization and subtractive hybridization. Sequence alignment and phylogenetic analysis of nucleic acid segments known to be among the most rapidly diverging in the organism are useful in determining the time the suspect sample diverged from its closest known relative, when time scales of nucleotide substitution are known. The use of these methods would be facilitated by precompiled databases of the target characteristics for available strains of potential bioterrorism species. In addition, molecular screens for tell-tale signs of genetic tampering can be employed.

The epidemiology toolbox for microbial forensics. R. C. SEEM (1) and H. Scherm (2). (1) Department of Plant Pathology, Cornell University, New York State Agricultural Experiment Station, Geneva, NY; (2) Department of Plant Pathology, University of Georgia, Athens, GA. Phytopathology 94:S124. Publication no. P-2004-0034-SSA.

Epidemiological tools are crucial for proactive and reconstructive microbial forensics. Proactive processes are used to identify when and where anomalous disease events might occur in the future. Examples of proactive methodologies include climate matching, used to estimate locations where a plant disease could become established, and trajectory analysis, used to plot long-distance transport of aerial pathogens. Reconstructive methodologies can be applied to identify anomalous disease events and trace them back to their original incidence, thus offering important clues about when and where the initial event occurred. Examples of epidemiological tools for reconstruction of a disease event include ground and remotely sensed disease and crop observations, survey and sampling plans, as well as models for disease spread, host plant development, and the analysis and interpolation of meteorological information. On the local scale, sporulating regions on a lesion or extent of plant necroses can be used to estimate time of initial symptom development; in addition, age-related susceptibility of host plants can shed light on the phenological stage of development at the time of infection. Regionally, reanalysis of archived weather forecasts can generate very detailed meteorological conditions preceding the identification of anomalous disease events.

***Pseudomonas syringae*: A model for forensic studies.** C. L. BENDER. Oklahoma State University, Stillwater, OK. Phytopathology 94:S124. Publication no. P-2004-0035-SSA.

An important aspect of microbial forensics is the rigorous detection of variation between related microbial strains and the use of variation to infer the origin, relationships, or transmission route of a particular isolate. Variation can be detected using a number of approaches including genome sequence polymorphisms, transcriptional profiling, differences in protein production, and metabolic profiling. These techniques can be used to study population structure, species evolution, and acquisition of virulence. With the completion of the genomic sequence, *P. syringae* pv. *tomato* DC3000 will be discussed as a model organism for forensic investigations in light of the vast information on epidemiology, genetic variation, and virulence in this pathogen.

The pathogen portal project – interoperability across data and tools for infectious disease research and development. B. SOBRAL. Virginia Bioinformatics Institute. Phytopathology 94:S124. Publication no. P-2004-0036-SSA.

Infectious diseases kill over 17 million people every year and cause significant economic damage to crop and animal systems that produce our food. Biological research tools have been industrialized to the point where it is now feasible to implement a systems approach toward understanding the host-pathogen-environment "disease triangle". Bioinformatics includes mathematical or computational (mathematical) biology science and Information Technology (hardware, network, software) infrastructure. Bioinformatics is one of the three main infrastructural components identified by diverse agencies as limiting. One main IT component needed for pathogenic microorganism research is a science portal (or computational "collaboratory"), especially one that integrated with computational science and tools. An infectious disease science portal stores and organizes biological characterization of known pathogens (and their near relatives) and provides what is needed to compute on that information. This resource would be a fundamental knowledge and decision-making tool for diverse federal and state agencies as well as for academic and other scientists working to improve our defenses against infectious diseases. VBI's PathPort Project was funded in 2002 by DoD, based on a five-year vision. First year funding was directed primarily to data acquisition and IT infrastructure development. The molecular data started at the genomic level and focused initially on the microbial component of the disease triangle. Second year funding is being directed at transcriptional profiling, with out years focused on protein and metabolite profiling as well as, ultimately, GIS and clinical data integration.

Species Concept in Host Pathogen Interactions, What Is Its Effect on Breeding for Resistance?

Introduction: Species concept in a commercial breeding program? M. R. MILES. USDA-ARS. Phytopathology 94:S124. Publication no. P-2004-0037-SSA.

As a plant pathologist working in the seed industry the changes in species names was always carefully monitered. Any change needed to be evaluated as to it effect on the screening and breeding programs as well as how information was presented to others in the company. This symposium was designed to look at a few cases where host or pathogen species concept has been shown to play roles in host resistance management.

Host gene-pool specialization and species concepts in *Uromyces appendiculatus* and other pathogens of common bean. M. A. PASTOR-CORRALES (1), J. R. Steadman (2), and M. C. Aime (1). (1) USDA-ARS, Beltsville, MD; (2) University of Nebraska-Lincoln. Phytopathology 94:S124. Publication no. P-2004-0038-SSA.

We have assessed genotypic diversity in a global collection of isolates of *Uromyces appendiculatus*, causal agent of rust of common bean, using both molecular markers and virulence assays. Phenotypic and genotypic results were congruent, indicating the existence of two different pathogen gene pools that corresponded to the Andean and Middle American gene pools of the host. Results from both virulence assays and molecular genotyping also reveal that the *U. appendiculatus* Andean gene pool is less diverse and has greater host-specificity compared to the Middle American gene pool. From these results, we suggest that *U. appendiculatus* has specialized on each of the two different *Phaseolus vulgaris* host gene pools. Characterization of pathotypic and genetic variation of *Colletotrichum lindemuthianum* and *Phaeoisariopsis griseola*, the causal agents of bean anthracnose and angular leaf spot respectively, also reveals two groups of isolates corresponding to the two common bean gene host pools and demonstrating a host gene pool specialization.

From one species to many or many species to one? – The case of begomoviruses. J. K. BROWN. Univ. Arizona. Phytopathology 94:S124. Publication no. P-2004-0039-SSA.

Only recently have begomoviruses (genus, Begomovirus; family, Geminiviridae) been recognized as notable pathogens, worldwide. Begomoviruses are small, circular, ssDNA plant viruses, which are transmitted by members of the whitefly Bemisia tabaci complex, upon which they depend entirely for their dispersal. Based on extant begomoviral genome sequences, new and emerging viruses have diversified, resulting in more fit genotypes and the extinction of others. The means by which begomoviruses adapt and speciate have only recently been scrutinized genetically, however, diversification occurs through multiple mechanisms, including mutation, intermolecular recombination, and potentially chromosomal reassortment. The discovery of satellite DNAs associated with certain begomoviruses also influences diversification and speciation. A working begomoviral 'species' definition must therefore contemplate the outcome of a potentially complex diversification pathway yielding variously complex extant population structures, which are at times confounding to the less forgiving criteria employed for their classification in a species framework.

***Puccinia recondita*, leaf rust of cereal and grasses: Who's who?** L. J. SZABO (1), Y. Anikster (2), and J. Markova (3). (1) USDA ARS Cereal Disease Laboratory, Department of Plant Pathology, University of Minnesota, St. Paul, MN; (2) Institute for Cereal Crop Improvement, Tel Aviv University, Ramat Aviv, Israel; (3) Charles University, Prague, Czech Republic. Phytopathology 94:S125. Publication no. P-2004-0040-SSA.

Leaf rust of cereal and grasses was first described almost two hundred years ago and the taxonomy of this rust has been in flux for almost as long. The current taxonomic classification of *Puccinia recondita* is based primarily on spore morphology, with *formae speciales* subdivisions based on the host. The host range of *P. recondita* includes 24 genera of telial hosts and 21 genera of aecial hosts, suggesting that *P. recondita* is actually a species complex, rather than a single species. However, until recently, the tools were not available to reliably differentiate members of this complex. DNA sequence analysis of the nuclear ribosomal RNA gene repeat was used to investigate this complex. Phylogenetic analysis of the current DNA sequence data indicated that there are at least 13 distinct species. Recent morphological and biological data, where available, support the DNA analysis. Elucidating the phylogenetic relationships and therefore the correct taxonomic classification will be essential to understanding the biology of this complex of cereal and grass leaf rusts.

Does the species concept matter in breeding for resistance? The maize gray leaf spot experience. M. CARSON. USDA-ARS, University of Minnesota, St. Paul, MN. Phytopathology 94:S125. Publication no. P-2004-0041-SSA.

Gray leaf spot (GLS) has become a serious disease of maize throughout the U. S. and much of the world. GLS resistant hybrids have been developed commercially based on field screening. At least three species of *Cercospora* have been associated with GLS; two sibling species of *C. zeae-maydis* and *C. sorghi* var. *maydis*. When isolates of these three species were tested for pathogenicity on a set of maize hybrids in field trials, there was no significant difference in pathogenicity between the two sibling species of *C. zeae-maydis*. There were no hybrid × sibling species interactions, as hybrids resistant to one species were resistant to the other. There were, however, significant isolate × hybrid interactions within each of the two sibling species, which resulted from significant variation in aggressiveness within each species. Less aggressive isolates were less effective in discriminating levels of resistance among the hybrids than more aggressive isolates. Isolates of *C. sorghi* var. *maydis* tested were not pathogenic, but appeared to be effective saprophytes.

Epidemiology/Ecology/Environmental Plant Pathology

Active Management of Soil Microorganisms for Plant Root Disease Control

Disease suppressive soils: Mechanisms and indicators based on microbial responses and soil chemical/physical properties. R. P. DICK (1), M. C. Cespedes Leon (2), N. Ochiai (3), A. Stone (4), M. Powelson (5), and F. Crow (5). (1) School of Natural Resources, Ohio State University; (2) National Institute of Agricultural Research (INIA-Quilamapu), Chile; (3) Department of Crop and Soil Science, Oregon State University, Corvallis, OR; (4) Department of Horticulture, Oregon State University, Corvallis, OR; (5) Department of Botany and Plant Pathology, Oregon State University, Corvallis, OR. Phytopathology 94:S125. Publication no. P-2004-0042-SSA.

Organic amendments can suppress plant diseases. However, mechanisms and soil indicators of suppression are not always well understood. We focused on soil microbial (enzyme activity, biomass, respiration), chemical (pH, nutrients) and physical (aggregation) properties that could be easily adopted for research and practical disease suppression applications. We studied suppression of common root rot (*Aphanomyes euteiches*) on snap beans with paper-mill residual by-products (composted vs. non-composted at 0, 22 or 33 dry Mg ha^{-1}) and *Verticillium dahliae* on potato with cover crops (0, 6, 12 or 24 Mg ha^{-1}) at field sites in Wisconsin (3 yrs of applications) and Oregon (1 yr applications), respectively. For both experiments organic amendment type and rates significantly affected disease ratings. For the longer experiment (WI), root rot was strongly, negatively correlated with total C and arylsulfatase (FDA showing no correlation). However, the OR experiment with only 6 months of field incubation of cover crop residues and subsequent potato planting/disease ratings; pH and either microbial biomass or respiration were most closely related to *V. dahliae* incidence. Implications of this preliminary disease suppression research and other aspects of organic amendment type, mechanisms, and soil quality indicators will be discussed.

Crop rotation and amendment effects on soil microbial communities and soilborne diseases. R. P. LARKIN. USDA-ARS, Orono, ME. Phytopathology 94:S125. Publication no. P-2004-0043-SSA.

Soil microbial ecology is greatly influenced by the type and quantity of plant material present. Different crop plants, as rotation, cover, or green manure crops, may be used to shape or alter microbial communities to increase primary crop production and reduce soilborne diseases. Crop rotations can reduce soilborne diseases by interrupting the host-pathogen cycle, stimulating microbial activity, diversity, and beneficial soil microorganisms, and by direct inhibition of plant pathogens. The role of crop rotations in the manipulation of soil microbial communities for the development of sustainable disease suppression will be discussed using examples from recent research on potato cropping systems. Different rotation crops result in distinctly different microbial community characteristics, based on population dynamics, substrate utilization, and fatty acid profiles, and may be related to effects on soilborne diseases. More research on the relationships and interactions of cropping practices on soil microorganisms and their effects on crop health is needed for the development of efficient, sustainable production systems.

Significance of host genotype in exploitation of resident disease suppressive soil microbial communities. M. MAZZOLA (1), Y.-H. Gu (1), D. L. Funnell (2), M. F. Cohen (1), and J. M. Raaijmakers (3). (1) USDA-ARS, Wenatchee, WA; (2) USDA-ARS, Lincoln, NE; (3) Wageningen University, The Netherlands. Phytopathology 94:S125. Publication no. P-2004-0044-SSA.

Impact of plant genotype on plant-microbe interactions has been widely examined in the realm of plant pathogenic organisms. Consideration of host specificity in interactions between plants and resident non-symbiotic plant-beneficial soil microorganisms has received decidedly less attention. The capacity of wheat to modify composition of the fluorescent pseudomonad population resident to orchard soils was found to vary in a cultivar-specific manner, and was associated with development of soil suppressiveness toward *Rhizoctonia solani* AG-5 and AG-8, pathogens of apple and wheat, respectively. Wheat genotypes also varied in the capacity to select, both qualitatively and quantitatively, for resident populations of 2,4-DAPG-producing fluorescent *Pseudomonas* spp., which have a pivotal role in the development of take-all decline. Apple rootstocks varied in the ability to support antagonistic pseudomonad genotypes enhanced through wheat cultivation and populations of resident *Streptomyces* spp. promoted by canola seed meal amendment. These findings implicate the importance of host genotype in optimizing use of resident soil microorganisms for disease suppression.

Management of soil microorganisms for the control of *Phytophthora* root rot. J. A. MENGE and V. McDonald. University of California, Riverside, CA. Phytopathology 94:S125. Publication no. P-2004-0045-SSA.

We have explored several microbial management strategies for the control of *Phytophthora* root rot of avocado and citrus. Mulching has increased the growth and yield of avocado in *Phytophthora*-infested soil by as much as 117% and 13%, respectively. Organisms active in mulch degradation secrete cellulase and laminarinase enzymes. Since the cell walls of *Phytophthora* contain cellulose and laminarin, *Phytophthora* is killed during the mulch decomposition process. Soils suppressive to *Phytophthora* have been reported by many authors, and these soils provide examples of how microorganisms

can efficiently control disease under natural conditions. Many biocontrol organisms have been reported to inhibit *Phytophthora* and reduce root rot caused by *Phytophthora*, however, none of these organisms has proven consistently effective under field conditions. Rhizosphere inhabiting microorganisms, general fungal parasites, antagonists and competitors all have inherent drawbacks which restrict their success as biocontrol agents against *Phytophthora*. However, obligate parasites, similar to those used successfully by entomologists for the control of insect pests, show promise, although they are notoriously difficult to grow. Foreign exploration for biocontrol agents which inhabit the native territory of *Phytophthora*, is another idea which can be gleaned from successful entomology biocontrol efforts. Finally an inundative strategy of continuous application of biocontrol agents through irrigation water eliminates many of the problems with current biological control efforts and has shown promise under field conditions.

Effect of cover crop decomposition on soil microbial and plant pathogen dynamics. N. GRUNWALD. USDA-ARS Vegetable and Forage Crop Research Unit, Prosser, WA. Phytopathology 94:S126. Publication no. P-2004-0046-SSA.

Cover crops are receiving increasing attention for their benefits in improving soil fertility and increasing suppressiveness to soilborne pathogens. Stages of oat-vetch cover crop decomposition were characterized over time in terms of carbon and nitrogen cycling, microbial activity and damping-off pathogen dynamics in organically and conventionally managed soils. Both field and controlled incubation experiments were conducted. Disease incidence and relative growth of *Pythium aphanidermatum* and *Rhizoctonia solani* were measured in growthchamber assays, in vitro growth tests, and field experiments. Incorporated residues led to immediate significant increases in biomass of microorganisms, microbial activity, and C/N ratios of debris. No significant differences were detected between the conventional and organic farming systems with respect to either relative growth or disease incidence. Total C and N content of debris and NH4-N content explain observed *P. aphanidermatum* dynamics most consistently. Cover crop decomposition is a very dynamic process and affects the soil microbial community and plant pathogen populations in complex ways.

Role and use of arbuscular mycorrhizae in root disease management. R. G. LINDERMAN. USDA-ARS Horticultural Crops Research Laboratory, Corvallis, OR. Phytopathology 94:S126. Publication no. P-2004-0047-SSA.

Soilborne pathogens causing root diseases must compete, prior to penetration of root tissue, with rhizosphere soil bacteria, actinomycetes, fauna, and fungi, including those that form mycorrhizae. Mycorrhiza formation causes physiological changes and direct or indirect rhizosphere microbial shifts that can affect the behavior of pathogens. Some reports indicate that mycorrhizae can induce some increased resistance in tissues, but a more likely explanation for root disease reduction is increased microbial antagonism in the mycorrhizosphere. Time to establish the symbiotic relationship before pathogen ingress is required for disease suppression to occur, and the background soil or growth medium must have sufficient antagonists to increase in association with mycorrhizae. Organic amendments, such as composts, could increase the level of antagonists in the background soil. However, inoculation of transplants with AM fungi and antagonistic associates prior to field planting could also result in sufficient antagonism to suppress root pathogens.

Pavich Family Farms: Trials and tribulations of an organic table grape grower. S. PAVICH. Pavich Agricultural Consulting. Phytopathology 94:S126. Publication no. P-2004-0048-SSA.

Pavich Family Farms has 33 years of farming history. Beginning with father's table grape vineyard in Delano Ca., the family farm grew from 250 acres to 4,400 acres. The Paviches developed the first large scale organic CCOF certified produce line, with 75 different produce products in health food stores and supermarkets in the United States and 50 million dollars in annual sales. We were also the first company to contract with small organic family farmers, at one point having 75 different farms in 6 countries. During the 80's and 90's, I sat on many boards, like the National Organic Standards Board at the USDA. My early education at Fresno State in the late 60's in Ag. sciences left me ill prepared to face the challenges of Organic Farming. What was taught in college was basic NPK agriculture. What was being taught by the early organic gurus was that if you apply tons of organic matter into the soil, that some magical balance would happen and soils that were conducive to disease would somehow change and become suppressive. I began composting to see if indeed I could create this ideal soil, and while many lessons were learned on what not to do, most of what was going on in the soil was still a big mystery. This session will address briefly some of my experiences over these 33 years farming organically, and my experience field testing hundreds of microbial products during this time.

Coevolutionary Processes of Introduced Pathogens and Hosts in Natural Ecosystems

Implications of molecular evidence on the evolution of *Melampsora* and other rust fungi. M. H. PEI, C. Bayon, and C. Ruiz. Rothamsted Research, Harpenden, Hertfordshire AL5 2JQ, UK. Phytopathology 94:S126. Publication no. P-2004-0049-SSA.

The rust fungi (*Uredinales*) are one of the largest groups of fungi. They thrive only on living tissues of plants, produce up to five different types of spores and often infect taxonomically very different plants to complete their life-cycle. Over more than a century, the evolution of rust fungi has been a fascinating subject of speculation among mycologists. We examined the evolutionary relationships among *Melamspora* and related groups of rust fungi using ribosomal DNA (rDNA) sequences from *Melampsora* on *Salicaceae* and the sequence information from other rusts available in GenBank databases. The rDNA sequence data suggested that multi-spore form life-cycle is likely to have evolved at very early stages of rust evolution. It appears that *Melampsora* split from ancestors of other rust genera at early stages of evolution of rust fungi. The split between the rusts having sessile teliospores (traditionally grouped in *Melampsoraceae*) and those having pedicelled teliospores (traditionally in *Pucciniaceae*) may have occurred before the split between *Melampsora* and other rusts having sessile teliospores. Some view points on the evolution of rust fungi, such as the ancestry, host specificity and the timing of divergence among different groups, were discussed in relation to the available molecular evidence.

A phylogeographical history of *Mycosphaerella graminicola* on wheat. B. A. MCDONALD and S. Banke. Institute of Plant Sciences, Swiss Federal Institute of Technology (ETH), Zuerich, Switzerland. Phytopathology 94:S126. Publication no. P-2004-0050-SSA.

Where do plant pathogens originate and how do they evolve? What processes define their evolutionary past and possible evolutionary future? Can we predict future evolutionary trajectories by understanding past evolutionary history? These are questions that lie at the heart of this talk. Answers to these questions can orient the search for sources of resistance in agricultural crops, because host resistance is most likely to occur at the center of origin of the pathogen. An understanding of the paths of recent pathogen movement (i.e. gene and genotype flow) can point to weak links in quarantine systems. We combined microsatellite and DNA sequence data from neutral RFLP loci and housekeeping genes with phylogenetic and coalescent analysis to define phylogenetic relationships and paths of genetic exchange among 384 isolates of the wheat pathogen *Mycosphaerella graminicola* originating from 14 populations on four continents. Though this talk will focus on *M. graminicola*, the coalescent approaches should be applicable to any plant or animal pathogen, and can be applied in the context of biosecurity to determine sources of newly introduced pathogen populations, whether through increasing international travel and commerce, or through bioterrorism.

Epidemiology meets genetics: The evolution of virulence in a natural plant-pathogen association. P. H. THRALL and J. J. Burdon. CSIRO Plant Industry, GPO Box 1600, Canberra ACT 2601, Australia. Phytopathology 94:S126. Publication no. P-2004-0051-SSA.

Genetic variation in hosts and pathogens has long been accepted as a crucial factor influencing disease epidemiology in human, plant and animal systems. However, broad understanding of the ecological and evolutionary dynamics of disease depends fundamentally on recognizing that such interactions occur in heterogeneous environments that can vary dramatically in space and time, and that this heterogeneity can have major impacts on maintenance of polymorphisms in relevant host and pathogen genes. Over more than a decade we have used the genetically well-understood interaction between native Australian flax (*Linum marginale*) and the rust pathogen (*Melampsora lini*) to investigate the interplay of host and pathogen genetics and demography across multiple populations subject to varying degrees of selective pressure, and through this, to elucidate the role of coevolutionary interactions in shaping the long-term dynamics of host-pathogen associations. Complementary to these empirical studies has been the development of a series of spatially explicit models which have been used to explicitly investigate how both epidemiological and coevolutionary outcomes depend on host and pathogen life-history. Here, we summarise results from these studies, and use

these to illustrate how the interplay of demographic and genetic processes influences the coevolution of host resistance and pathogen infectivity at spatial scales ranging from single populations to metapopulations and beyond. We conclude by discussing the implications of our findings for understanding epidemiological and evolutionary outcomes in exotic host-pathogen interactions.

Inferring patterns of migration and gene flow in introduced populations of plant pathogens using the chickpea pathogen *Ascochyta rabiei* as a model. T. L. PEEVER. Dept. of Plant Pathology, Washington State University, Pullman, WA 99164-6430. Phytopathology 94:S127. Publication no. P-2004-0052-SSA.

Inferring patterns of migration and gene flow among populations of introduced plant pathogens is problematic because it is rarely possible to separate historical association from contemporary gene flow. Migrations of plant pathogens into agricultural regions previously free of the disease are recent events relative to the co-evolutionary history of host and pathogen. Populations are likely to represent a subset of variation found in the center of origin and are not expected to be in equilibrium. Migration events and sources of introductions can be inferred with precision using highly polymorphic molecular markers applied to samples preserved from the original introductions and from the region where plant and pathogen have co-evolved. An extensive worldwide collection of isolates and a genetically characterized set of molecular markers (microsatellites and mating type) are available for *Ascochyta rabiei*, the causal agent of Ascochyta blight of chickpea. These markers, plus isolates from the putative center of origin of both plant and fungus, are being used to infer sources of recent introductions in the US Pacific Northwest and California. Isolates sampled over a six-year period from a single California population were used to test the hypothesis that the mating system of the fungus has shifted from asexual to sexual concomitant with the introduction of the alternate mating type. The implications of this mating system shift for pathogen biology and evolution and the epidemiology of Ascochyta blight will be discussed.

Exotic and native rust pathosystems: A population genomics approach. R. C. HAMELIN. Natural Resources Canada, Laurentian Forestry Center, 1055 du P.E.P.S., Quebec, Canada G1V 4C7. Phytopathology 94:S127. Publication no. P-2004-0053-SSA.

Rusts pathogens cause some of the most severe tree diseases. In order to better understand whether or not native rusts have adaptive advantage over invasive ones, we are searching for expressed sequence tag polymorphisms (ESTp) in genomic DNA of several native and invasive rusts species and generating genomic sequence data at the population level. Our findings show that white pine blister rust has a very narrow genetic basis in eastern North America, but an even narrower basis in western North America. Nucleotide diversity, Pi, was three times as high in eastern than in western populations. However, high ratios of non-synonymous/synonymous mutations were found at important loci, such as a cytochrome P450 which is involved in phytoalexin detoxification in some pathosystems. By contrast, the native poplar leaf rust caused by *Melampsora medusae* has a very high genomic diversity with multiple alleles per locus in North America, but a very narrow genetic basis in Europe where it was introduced. In North America, *M. medusae* covers the range of distribution of poplars over 30 degrees of latitude, but in Europe it is mostly limited to southern regions. We are currently testing whether such restrictions in distribution are due to the absence of essential genetic determinants.

Implications from deep phylogeographical histories on pathosystem endemism. G. I. MCDONALD. USDA Forest Service, RMRS, Moscow, ID. Phytopathology 94:S127. Publication no. P-2004-0054-SSA.

Ancient mosaics of plant, animal, and fungal communities dispersed across the landscape can be detected only by reading evolutionary history recorded in genomes. Knowledge about the distribution and nature of these mosaics in relation to pathosystem endemism (PE) is essential for sustaining effective pest management. Damage to white pines by blister rust (BR) in North America (NA) has ranged from low to high, and some heavily impacted populations have recovered rapidly. Geographic distribution of resistance genes in NA varies among subpopulations delineated by isozymes and maternally inherited haplotypes (MIH). BR ecology discriminates subpopulations in Asia. Together these observations raise questions about the conceptualization of PE. Congruent *Cronartium* and *Pinus* phylogenies supports the Fahrenholtz Rule and point to BR's origin in NA. "Suture" zones (SZ) revealed by MIH may provide a key to understanding PE. Some SZ show 2 to 3 million years of divergence that has persisted through episodes of climate shifts and hemispherical glaciations. Retarded genetic homogenization (RGH) associated with SZ indicates that hidden genetic structures are retained during repeated migrations across the landscape. Retention of interspecific crossing compatibility, intraspecific pathosystems, and other traits may indicate RGH tracing back to the breakup of Pangea. Boundaries identified by MIH and dwarf mistletoe distributions reveal potential NA SZ. These SZ may indicate where complex interactions among refugia and recolonization dynamics have disrupted pathosystems.

Food Safety as Influenced by *Phyllosphere microflora*

Putting science to work: Separating the possible from the plausible. T. SUSLOW. University of California, Davis. Phytopathology 94:S127. Publication no. P-2004-0055-SSA.

Food-borne pathogens are a major cause of human morbidity and mortality worldwide. In the United States acute gastroenteritis is the second most common household illness and an estimated 76 million food-related illnesses occur each year. The cumulative cost of medical expenses, lost wages, and productivity; approaches $10-14 million annually. Producers, shippers, processors, as well as foodservice, and retail handlers have been investing in the design and implementation of Good Agricultural Practices and comprehensive food safety systems for over eight years. Despite this effort, recent, large outbreaks of salmonellosis, *E. coli* O157:H7 and hepatitis virus A that have resulted in multistate illnesses and some deaths punctuate the fact that there are gaps existing scientific knowledge of ag-environmental ecology of human pathogens, plant:pathoghen interactions, and human pathogen:associated microflora interactions. Failures to apply existing knowledge in these areas are another contributing factor. Examples from case studies, on-farm surveys, and environmental persistence research will be presented. From a research-to-application perspective, this presentation will explore the question "What does Ready-to-Eat" mean for fresh horticultural products.

Going from compost to compost tea: Weighing plant health benefits against human pathogen uncertainties. S. SCHEUERELL. Oregon State University. Phytopathology 94:S127. Publication no. P-2004-0056-SSA.

Using biological wastes as agricultural amendments has a number of benefits and risks to both plant and human health. Properly composting wastes has traditionally been viewed as a solution to gain the plant pathogen suppression and nutrient benefits from organic wastes, while minimizing potential risks, particularly reducing pathogenic organisms associated with waste materials. In recent years, agricultural producers have been producing compost tea from compost, water, and nutrient additives in order to transfer and multiply the microbial populations from compost into an aqueous phase that can be applied to plant surfaces and soils in ways not logistically or economically feasible with compost. This practice has generated both reports of plant disease control and the potential to increase residual populations of human pathogenic bacteria from the starting materials. The impact of various compost tea production practices on the benefits and risks of compost tea will be discussed in relation to the proposed recommendations of the National Organic Standards Board Compost Tea Task Force.

Use of non-composted bovine manure as fertilizer: An evaluation of vegetable contamination risk. S. INGHAM. University of Wisconsin, Madison. Phytopathology 94:S127. Publication no. P-2004-0057-SSA.

Over the past 5 years, we have tested the validity of the National Organic Program (NOP) requirement for a >120-day interval between application of non-composted manure and harvesting of vegetables grown in manure-fertilized soil. Results of initial studies using soil beds in a controlled environment chamber suggested that the NOP limit was overly conservative. Field surveys of Wisconsin soils fertilized with non-composted bovine manure supported this suggestion. In spring, 2003, non-composted bovine manure was applied on 9.3 m2 plots at three Wisconsin sites (loamy sand, silt loam, and silty clay loam) prior to spring/summer planting of carrots, radishes, and lettuce. No chemical pesticides or herbicides were applied to the plots throughout the study. Soil and washed (30 s under running tap water) vegetables were analyzed for indigenous *Escherichia coli*. Within 90 days, E. coli in manure-fertilized soil generally decreased by about 3 log CFU/g from initial levels of 4.1 – 4.4 log CFU/g, with fastest die-off in the loamy sand. However, low levels of *E. coli* generally persisted in manure-fertilized soil for over 100 days and were detected in enriched soil from all three sites 132-168 days after manure application. For carrots and lettuce, at least one enrich-

ment-negative sample was obtained 100-119 days after manure application for 56 and 84% of treatments, respectively. The current >120-day limit provided an even greater likelihood of not detecting *E. coli* on carrots (>1 enrichment-negative result for 84% of treatments). The rapid maturation of radishes prevented conclusive evaluation of either a 100 or 120-day application-to-harvest interval. Our 2003 results suggested that the absolute absence of *E. coli* on vegetables harvested from manure-fertilized Wisconsin soils may not be ensured solely by adherence to the NOP>120-d limit, but the safety of vegetables grown in these soils would not be unduly jeopardized by reducing the NOP requirement to >100 days. Other Good Agricultural Practices no doubt play an important role in ensuring that vegetables are safe to eat. In 2004 studies, we are using larger plots and mechanized manure spreading, tilling, and cultivation, and we are applying pesticide/herbicide when necessary. Preliminary results from the 2004 studies will be presented and compared to those from 2003.

Biofilms and other strategies exploited by salmonella and friends on plants. J. BARAK. USDA-ARS, Albany, CA. Phytopathology 94:S128. Publication no. P-2004-0058-SSA.

Salmonella enterica, *Escherichia coli* and *Erwinia chrysanthemi* are related bacteria with interconnected roles in food safety and quality. *S. enterica* and *E. coli* are human pathogens and have caused foodborne outbreaks associated with produce. *E. chrysanthemi* is a plant pathogen that causes soft rot diseases on produce and has been associated with a higher incidence of human pathogens on produce. Although *S. enterica* and *E. coli* infect animals and *E. chrysanthemi* infects plants, these species share environmental niches such as water, fresh produce, and insects, and they carry homologous virulence genes. We have identified culture conditions that differentially promote biofilm formation on glass in each species. Our results suggest that *E. chrysanthemi*, *E. coli*, and *S. enterica* use homologous genes for biofilm formation and the presence of *E. chrysanthemi* helps induce *E. coli* and *S. enterica* biofilm formation on glass under conditions in which the human pathogens do not form biofilms in pure culture. Attachment and multicellular behavior are essential first steps in enterobacterial biofilm formation and recent reports suggest that multicellular behavior requires production of cellulose and protein filaments. Our research shows that both human and plant pathogens use the same molecular player for multicellular behavior on abiotic surfaces. Building on this work, we have determined mechanisms required for human pathogens in association with plants. Our earlier work revealed differences between the human pathogens, *S. enterica* can adhere to plant tissue and is impossible to remove from fresh produce without significantly reducing its quality and *E. coli* O157:H7 can easily be removed. Our research reveals the importance of csgD and rpoS in initial attachment to plants for *S. enterica*. CsgD activates both the expression of curli and production of cellulose. We hypothesize the poor attachment by *E. coli* O157:H7 to plants may be because they rarely produce curli, due to mutations in the csgD promoter.

Fitness of *Salmonella enterica* in the phyllosphere. M. BRANDL. USDA-ARS, Albany, CA. Phytopathology 94:S128. Publication no. P-2004-0059-SSA.

Recent outbreaks of food-borne illness linked to fresh produce have raised concerns regarding the ability of human enteric pathogens to grow and survive on plants in the pre-harvest environment. *Salmonella enterica* serovar Thompson was associated with an outbreak from cilantro in California. We have investigated the fitness of *S. thompson* in the cilantro phyllosphere and its interaction with common soft-rot plant pathogens such *Erwinia chrysanthemi* and *Pseudomonas viridiflava*. Our studies demonstrate that *S. thompson* reached higher population sizes and was more competitive on cilantro leaves at 30°C than at 24°C, and that it recovered from short periods of dry conditions on plants to the same extent as common bacterial epiphytes. Microscopy of GFP-labeled *S. thompson* cells on leaves combined with image analysis revealed that 70% of the cells were located in large aggregates, and that quorum-sensing signaling via autoinducer-2 was not involved in the formation of aggregates, nor in the fitness of *S. thompson* on plants. Unlike with *P. viridiflava*, *S. thompson* population sizes were highly correlated with those of *E. chrysanthemi* and its various virulence mutants after their co-inoculation onto cilantro plants. *S. thompson* and *E. chrysanthemi* formed large mixed aggregates on cilantro leaves. The role of cross-talk via autoinducer-2 in the interactions between these two species in the phyllosphere will be discussed.

Influence of indigenous bacteria on survival of human pathogens on plants. C. POZA-CARRIÓN and S. E. Lindow. University of California, Berkley. Phytopathology 94:S128. Publication no. P-2004-0060-SSA.

Although generally associated with consumption of meat products, numerous food-borne diseases caused by *Salmonella enterica* serotypes and *Escherichia coli* O157:H7 have been associated with contaminated fruits and vegetables in the United States, as well as in many other countries. The surfaces of plants are normally colonized by large numbers of bacteria, called epiphytes, which include plant pathogens and other saprophytic and beneficial bacteria. Preliminary studies have shown that while *E. coli* and *Salmonella* strains can grow on plants under conditions of continuous free moisture, they are hypersensitive to the stress of dry leaf surfaces compared to other bacteria more commonly found on plants, indicating that stress tolerance is a determining factor in their epiphytic colonization. Extrinsic factors such as patterns of colonization of plants also have a great impact on the survival of bacteria on leaves. Cells of *Pseudomonas syringae* in aggregates on leaves were much more stress tolerant than solitary cells and the survival of immigrants of *P. syringae* to leaves was greatly enhanced if cells were deposited onto pre-existing aggregates of a variety of bacteria on plants. The survival of *Salmonella enterica* subsp. *enterica* serovar Montevideo and serovar Enteritidis and *Escherichia coli* O157:H7 inoculated onto lettuce and cilantro leaves was often strongly influenced by the number and strain identity of indigenous epiphytic bacteria preexisting on a leaf. We will also discuss fluorescence microscopic studies that address the role of bacterial cell aggregates in survival of such strains at small spatial scales.

Infective dose of enteric pathogens: Influence of post harvest processing and storage practices. A. BHAGWAT. USDA-ARS, Beltsville, MD. Phytopathology 94:S128. Publication no. P-2004-0061-SSA.

The complex nature of today's food distribution chain creates opportunities for contamination of fresh-cut fruits and vegetables. Contamination of some fresh produce (sprouts, apple cider) is potentially more serious than contamination of animal products because these foods may be more likely to be consumed without further processing treatments that would kill pathogenic microbes. The number of bacteria consumed in contaminated food can vary considerably, as does the infective dose (ID) of enteric pathogens. The ID of *Shigella* and diarrheagenic *Escherichia coli* strains (highly acid-resistant), and ID of *Salmonella* and *Vibrio* strains (mildly acid-resistant) roughly correlates with their ability to survive stomach acidity. Dose response is an outcome of interdependent effects among organisms, host, and food vehicle. Although there have been numerous studies of the mechanisms of attachment of bacteria to human and animal cells, virtually nothing is known about the attachment of bacteria to foods and food contact surfaces. Even much less is known about how these processes contribute towards bacterial virulence and their ID. We found that attachment of *Salmonella* strains to fresh-cut produce surface played a critical role in increasing their acid tolerance. Other specific examples of post-harvest factors that influence ID (acid-tolerance) and survival of diarrheagenic *E. coli* and *Shigella* strains are: (a) controlled atmosphere packaging and storage conditions, (b) availability of glutamate and arginine from the fresh-cut produce, and (c) pH of the anti-browning wash treatment solutions. Genetic analysis of 82 foodborne outbreak-associated pathogenic *E. coli* strains (isolated from 34 countries and 23 states within the U.S.A.) revealed 36% strains carried mutation(s) in the alternative transcription factor gene, *rpoS*. Thus, given the plasticity of microbial evolution, it is important to determine if certain food processing practices are exerting selective pressures for the emergence of more competent pathogenic strains.

Soil Health and Nematodes

Nematode diversity in soil ecosystems. G. W. YEATES. Landcare Research, Palmerston North, New Zealand. Phytopathology 94:S128. Publication no. P-2004-0062-SSA.

Soil nematode assemblages typically contain over a million individuals m^{-2}, representing 30-200 species. A series of examples show that: 1. Economic crop loss may results from large populations of a single plant-feeding species (e.g., *Heterodera schachtii*), a combination of species acting in series (e.g., *Heterodera*, *Meloidogyne*, *Pratylenchus*), or relatively low numbers of a single species (e.g., *Longidorus*); 2. Bacterial-feeding, fungal-feeding, predacious, omnivorous and plant-associated nematode functional groups are also each commonly represented by many species; 3. While recent work indicates many Tylenchidae to be fungal-feeding, still much remains to be determined about nematode feeding habits and resource use; 4. A range of studies has shown that grazing by nematodes on soil microflora (bacteria, fungi) and microfauna (nematodes, rotifers, enchytraeids etc) can increase turnover of populations in the soil, enhancing nutrient cycling and, potentially, reduce stresses on plants; 5. Nematodes interact with other soil biota; protozoa and rotifers also feed on bacteria; tardigrades, mites and fungi prey on nematodes;

6. There are gaps in information on differences in nematode biology that underlie population changes (e.g., moisture). It is believed that diversity within each nematode functional group is the key to resilience in the nematode assemblage and its ability to respond to environmental pressures, global change and agricultural management.

Role of nematodes in soil nutrient cycling. G. BIRD (1), R. Harwood (1), J. Sanchez (2), M. Berney (1), J. Smeenk (3), and J. Smith (1). (1) Michigan State University, East Lansing, MI 48824; (2) Oklahoma State University, Goodwell, OK 73939; (3) University of Alaska, Fairbanks, AK 99775. Phytopathology 94:S129. Publication no. P-2004-0063-SSA.

Taxa of the animal phylum Nematoda can be partitioned into guilds that colonize all ecosystems of our planet. As dissipative consumers, they transform and transport matter and energy throughout ecosystems. The excretory, secretory and necrotic products of species with soil inhabiting life cycle stages are directly involved in soil nutrient cycling. The objectives of this presentation are to: 1) describe a conceptual model of the role of nematodes in soil nutrient cycling: with special reference to carbon and nitrogen, 2) present an overview of the literature on the role of nematodes in soil nutrient cycling, 3) provide data on the nutrient mineralization potentials of a broad range of soils associated with Michigan agriculture, 4) demonstrate the impacts of agricultural management systems on associated nutrient mineralization potentials under Michigan growing conditions: including special reference to nematode community structure and 5) illustrate the impacts of selected biotic and abiotic ecosystem disturbances on nematode community structure and nutrient mineralization potentials. In conclusion, the presentation will use the conceptual model of the role of nematodes in soil nutrient cycling for the identification of knowledge gaps and recommendations for future research priorities.

Decoding the nature of a nematode suppressive soil. J. O. BECKER (1) and J. Borneman (2). Departments of (1) Nematology and (2) Plant Pathology, University of California, CA. Phytopathology 94:S129. Publication no. P-2004-0064-SSA.

In certain soils, survival, population development or activity of phytoparasitic nematodes are diminished by biological means despite otherwise conducive conditions. Analysis of such nematode suppressive soils may hold the key to advances in our understanding of biological management of these pests. However, little is known about the occurrence and distribution of nematode suppressive soils, causal agents, mode of action, and conditions that lead to or maintain suppressiveness. Our investigations have focused on a beet cyst nematode suppressive soil located at the Agricultural Operations near the UC Riverside campus. The biological nature of this soil suppression was demonstrated by targeting the responsible factors with biocides such as pesticides or heat. Amending conducive soils with small amounts of the suppressive soil or one of its components, nematode cysts, transferred the activity and established nematode population suppression within one growing season. Identification of the causal organisms was aided by exploiting selective sensitivity to chemical, physical and biological exclusion methods combined with in situ isolations, semi-selective media and media-independent microbial community analysis. Fungal parasitism of various life stages of *Heterodera schachtii* was found to be responsible for the nematode population suppression.

Soil microbial and nematode communities in organic and conventional farming systems. J. B. RISTAINO. North Carolina State University, Raleigh, NC. Phytopathology 94:S129. Publication no. P-2004-0065-SSA.

Research has been underway since 1997 in field plots located at the Horticultural Crops Research Station to determine whether organic fertility amendments are more suppressive to disease caused by *Sclerotium rolfsii* than soils with synthetic fertility amendments? In addition, we are interested in whether species diversity, functional diversity or the composition of the soil microflora is most closely related to disease suppressiveness? The experimental design was a replicated split-plot with tillage on bare-soil or tillage followed by surface-mulch with wheat straw as main plots, and organic soil amendments (cotton gin trash, animal manure or a rye vetch green manure) or synthetic fertilizer as subplots. In most years, cotton gin trash was highly suppressive to southern blight. Populations of bacterivorous nematodes mainly in the Rhabditidae and Cephalobidae, and fungivorous nematodes were greater after planting in soils amended with swine manure, composted cotton-gin trash, or rye-vetch, than in soils amended with synthetic fertilizer. Populations of Meloidogyne incognita in soil were not affected by soil amendments, but increased through time. Root-gall indices were lower in plots containing swine manure or cotton-gin trash than in those with synthetic fertilizer or rye-vetch in one year. Soils amended with cotton gin trash or animal manures contained higher numbers of bacteria and actinomycetes and species diversity was greater in these soils. Measures of impacts of the fertility amendments on disease, nematode communities, and abundance and diversity of soil microbial species communities will be discussed.

Effects of soil ecosystem management on nematode pests, nutrient cycling, and plant health. K.-H. WANG and R. McSorley. University of Florida, Gainesville, FL. Phytopathology 94:S129. Publication no. P-2004-0066-SSA.

Most agricultural practices affect soil ecosystems, but a challenging question is whether soil food webs can be deliberately managed to minimize problems from plant-parasitic nematodes. Ideally, we want to maintain a healthy soil food web that can 1) suppress nematode pests, 2) enhance soil nutrient cycling, 3) enhance natural enemies of plant-parasitic nematodes, and 4) improve plant health. Some conventional agricultural practices, including soil fumigation and herbicide application, can be destructive to nematode communities, suppressing natural enemies of nematodes and free-living nematodes involved in nutrient cycling. Organic agricultural practices, such as crop rotation, cover cropping, organic amendments, and biological control, not only reduced some key plant-parasitic nematodes, but also had multiple impacts on soil food webs. Organic amendments derived from cover crops, such as sunn hemp, stimulated bacterial and fungal-feeding nematodes as compared to inorganic fertilizer. However, this effect varied according to soil histories and C:N ratio of the residue. Succession of nematode communities occurred during the decomposition of residues and correlated with microbial biomass. Organic amendment also enhanced some natural enemies of nematodes, such as nematode-trapping fungi and omnivorous nematodes. The combined enhancement of nutrient recyclers and natural enemies of nematodes can contribute to the improvement of plant health. A key question remaining is whether the improvement of soil food webs by the above-mentioned approaches is sufficient to benefit farmers economically.

The Plant Pathologist's Toolkit for Responding to Crop Biosecurity Threats

The role of CSREES in responding to new and emerging agricultural pathogens. K. CARDWELL. National Program Leader, Plant Pathology, USDA, CSREES, Waterfront Bldg - Mail Stop 3153, 800 9th St SW, Washington, D.C. 20024. Phytopathology 94:S129. Publication no. P-2004-0067-SSA.

The U.S. Department of Agriculture has the mandate to promote and protect American agriculture. Because of the potential of terrorists or economic saboteurs to strike at American security and well-being through its vast food production systems, various branches of the USDA have shifted focus towards system protection. The Patriot Act of 2002 established the select agent list of plant and animal pathogens. The Agricultural Research Service (ARS) develops detection assays for these pathogens and the Animal Plant Health Inspection Service (APHIS) validates and deploys them. The Cooperative State Research, Education, and Extension Service (CSREES) coordinates with Universities across the country, bringing resources to bear at the State and local level. CSREES has a number of competitive integrated REE grant programs that are relevant to homeland security. Within the NRI, there are competitive programs for Plant and Animal Biosecurity, Biology of Plant-Microbe Associations, and Microbial Genomics Sequencing. A Critical and Emerging Issues grant provides seed money for start-up research on new pests and pathogens in the country. In addition to competitive grants, CSREES facilitates a number of nationally coordinated teams who work together to promote agricultural security. The National Plant Diagnostic Network (NPDN) connects diagnostic laboratories in Land Grant Universities across the country. The NPDN interfaces directly with the Regional IPM Centers and the Emergency Disaster Extension Network (EDEN), all of which are funded through CSREES. There are also very close liaisons with the Pesticide Safety Education Program (PSEP) and the national invasive species initiative. This symposium will explore ways to be a part of these important national networks and initiatives.

The role of NPDN in responding to new and emerging agricultural pathogens. J. P. STACK. Director, Great Plains Diagnostic Network, Kansas State University, Plant Science Center - Plant Path Dept, 4024 Throckmorton Hall, Manhattan, KS 66506-5502. Phytopathology 94:S129. Publication no. P-2004-0068-SSA.

A secure agricultural system requires rapid detection of outbreaks, accurate diagnoses of problems, and early response to minimize impact. Established in June 2002 by the United States Department of Agriculture (USDA) and the

Department of Homeland Security, the National Plant Diagnostic Network (NPDN) is a consortium of five regional networks and a national database designed to facilitate early detection and rapid diagnosis of outbreaks. Within each region is a regional diagnostic center that coordinates diagnostics, communications, and training. A program to improve the infrastructure of regional and state diagnostic laboratories has been implemented, including advanced diagnostics technology (e.g., PCR and real time PCR technology). Many state labs have been equipped with web-enabled microscopy to provide greater access to expertise regarding exotic pests and pathogens. For an efficient response during an outbreak, it is important to identify the positives and to clear the negatives. NPDN's system of parallel networked diagnostic laboratories provides surge capacity to assist in triage and preliminary diagnoses. The key to minimizing impact is a rapid response which is dependent upon effective and rapid communication. NPDN has established a secure and rapid communication system; vital to a secure agricultural system. It is essential that everyone who may pay a role during an outbreak understand their role and how it fits into an overall response plan. A national program has been established to develop and implement training exercises in all states that includes all cooperating agencies and institutions. Preparedness requires practice. A secure agricultural system requires skilled diagnosticians and first detectors. A program for training diagnosticians to keep current with advanced diagnostics and first detectors to raise awareness on high risk pests and pathogens as well as how to collect and submit a quality sample has been implemented.

What is on the horizon concerning new tools for detection of plant pathogens? R. R. MARTIN. USDA-ARS-HCRL, Corvallis, OR 97330. Phytopathology 94:S130. Publication no. P-2004-0069-SSA.

PCR has been the basis for the remarkable improvements in pathogen detection during the last several years. Multiplex, Real-time and Real-time multiplex PCR have reduced the cost and time to determine if a sample is infected with a known pathogen. For fungi, bacteria and phytoplasmas, primers that amplify ribosomal RNA or other conserved genes have been used as broad spectrum probes and are useful in detection of an unknown pathogen. Sequencing of the amplicons obtained in these general tests allows for the development of pathogen specific probes. Broad spectrum probes for viruses generally can be used to detect all known members of a genus or family. Obtaining sequence from isolated dsRNA can still be the most efficient means of identifying a new virus. The next major improvement will be the application of microarray technology for detection. Detection of viruses using microarrays may be possible without a PCR amplification step by using molecular beacons as trapping primers on the microarray. With the application of microarray technology it will be possible to develop chips for the simultaneous detection of all known pathogens of a crop or group of related crops.

Post-introduction mapping of new and emerging agricultural pathogens in real-time using GPS and GIS technologies. F. W. NUTTER, JR. Department of Plant Pathology, Iowa State University, Ames, IA 50011. Phytopathology 94:S130. Publication no. P-2004-0070-SSA.

One of the basic tenets of plant biosecurity is that the presence, actual and predicted distribution, disease intensity, and economic impacts of any yield-reducing factor(s) must be known. The coupling of diagnostic records originating from the National Plant Diagnostic Network coupled with global positioning systems (GPS) and geographic information systems (GIS), such as ESRI GIS software, have tremendous potential to enhance the value of diagnostic records. The real-time post-introduction monitoring of introduced agricultural pathogens in time and space by geospatially referencing and mapping diagnostic records provides a sound basis for policy-makers in their decisions concerning attempts at eradication and/or imposing quarantines, as well as facilitating timely disease management recommendations. These integrated technologies will also be important with regards to forensics and attribution.

Use of aerobiological information and meteorological trajectory analysis to predict and monitor spread of plant disease epidemics. C. E. MAIN, T. Keever, and G. J. Holmes. North American Plant Disease Forecast Center, North Carolina State University, Raleigh, NC 27695-0001. Phytopathology 94:S130. Publication no. P-2004-0071-SSA.

Epidemics caused by new and/or exotic airborne plant pathogens, such as mildews and rusts in the hands of bioterrorists could represent a serious threat to U.S. Agriculture. By applying atmospheric trajectory models and data to the aerobiological characteristics of fungal and bacterial pathogens, forecasting systems can be developed to monitor movement of propagules both into and throughout the United States. Such a system for the whole North American continent has been operative for nine years for tobacco blue mold (*Peronospora tabacina*) and six years for cucurbit downy mildew (*Pseudoperonospora cubensis*) from infected field sources in the Caribbean and Latin America. Daily forecasts are posted on an Internet site and available via a toll-free telephone number. More recently the potential for aerial movement of soybean rust spores (*Phakopsora pachyrhizi*) into the US is being monitored from known sources in South America and Africa. The technology and experience gained from using such forecasting systems since 1996 is available and should be considered for other important airborne diseases that pose bioterrorist potential.

Weather-based simulations of invasive plant pathogens. J. M. RUSSO (1), S. Isard (2), and R. Magarey (3). (1) ZedX, Inc., Bellefonte, PA 16823; (2) University of Illinois, Champaign, IL 61801; (3) North Carolina State University, Raleigh, NC 27695. Phytopathology 94:S130. Publication no. P-2004-0072-SSA.

Weather-based model simulations have flourished in recent years due to the steady advancement in information technologies for desktop computers. With their unprecedented storage, speed, and programs for analysis and visualization, today's systems allow for model simulations from the local to global scales. Like other weather-sensitive fields, plant pathology has taken advantage of the evolution of information technologies and computer power. This paper discusses the current state of weather-based simulations of invasive plant pathogens. It outlines possible weather-driven, modeling approaches for simulating disease development and movement. The paper places special emphasis on how users can access model output and interpret products derived from simulations in their decision making. The modeling approaches and user access are illustrated with examples, which include soybean rust (*Phakopsora pachyrhizi*).

Mitigating the post-introduction impacts of new and emerging agricultural pathogens. T. R. GOTTWALD (1) and D. T. Kaplan (2). (1) USDA-ARS-USHRL, Ft. Pierce, FL 34945; (2) USDA-APHIS-PPQ-CPHST, Raleigh, NC 27606. Phytopathology 94:S130. Publication no. P-2004-0073-SSA.

International travel and trade has greatly increased the introduction of plant pathogens. When introduced pathogens potentially have severe effects on valuable agrosystems and natural resources requiring state and federal regulatory responses. To ensure the most rapid and effective response to introduction, scientific studies are initiated to gain critical information for biologically sound pest mitigation. Responses to three pathogen introductions, citrus canker, plum pox virus, and *Ralstonia solanacearum*, are presented as case studies. Citrus canker, discovered in south Florida in 1994, resisted eradication attempts requiring a study that led to new regulations and eventually a 1900-ft law and development of a sentinel survey method used statewide. Over $500,000,000 has been spent to date and over 1.2 million trees removed to attempt to eradicate this pathogen. Plum pox virus was discovered in Pennsylvania in 1999 and Ontario, Canada in 2000, requiring several studies that have lead to improved state, national, and NAPPO survey methods and standards, eradication protocols, and rapid pathogen detection and differentiation. *R. solanacearum* race 3 biovar 2, a severe pathogen of solanaceous crops, was discovered in 2003 in imported geraniums, resulting in quarantines of commercial nurseries and studies to enhance sampling, detection, and disease suppression and eradication efforts.

Molecular/Cellular Plant-Microbe Interactions

Closteroviruses - *Citrus tristeza virus* Complex and Tristeza Diseases

Citrus tristeza diseases – a worldwide perspective. S. M. GARNSEY (1) and P. Moreno (2). (1) Univ. of Florida, CREC, Lake Alfred; (2) IVIA, Moncada, Valencia, Spain. Phytopathology 94:S130. Publication no. P-2004-0074-SSA.

Citrus tristeza virus (CTV) has had major impacts on citriculture. CTV-induced declines have destroyed millions of trees propagated on sour orange and forced major changes in citrus rootstocks, a process that created new disease problems. CTV-induced stem pitting diseases have chronically depressed citrus production in many countries and limit cultivar selection. Both host and virus isolate influence the wide array of symptoms observed. Viral sequence comparisons suggest diversity in origin as well as recombi-

nation and mutation. Diverse CTV isolates have been distributed in complex patterns via international movement of CTV-infected citrus plants and local spread by aphids and bud propagation. This process has yielded a continuing series of CTV problems and crop losses. Creating effective controls for the different CTV threats poses daunting technical challenges. While extensive new information on the biology, epidemiology and molecular properties of CTV has been generated recently through extensive international efforts, many properties of the virus crucial for understanding disease induction and formulating controls remain undetermined.

***Citrus tristeza virus* as a model and a tool – replication, assembly, interactions with the host, and transmission.** W. DAWSON, S. Gowda, T. Satyanarayana, A. Folimonov, S. Folimonova, M. Albiach, M. Ayllon, and C. Robertson. Dept. of Plant Pathology, CREC, Univ. of Florida, Lake Alfred, FL 33850. Phytopathology 94:S131. Publication no. P-2004-0075-SSA.

Citrus tristeza virus (CTV), a serious pathogen of citrus, is one of the most complex plant viruses. The approximately 20 kb single-stranded RNA genome encodes at least 19 proteins, ten of which are not needed for replication. Four of these gene products are needed for formation of virions, two are host range determinants, two have RNA silencing suppressor activity in addition to other functions, and two are unknown. Three genes can be deleted without noticeable effects on the ability of the virus to replicate and move in certain plants. The complex virion architecture consisting of major and minor coat proteins and at least two other accessory proteins allows specific interactions with its aphid vector. Hybrid viruses created from isolates of different phenotypes allow mapping of disease determinants as a first step in developing mild isolates to be used to cross protect trees from severe isolates of the virus. The ability to manipulate the genome of the virus also allows the development of virus-based transient expression vectors to examine gene expression in mature trees.

Variation in composition and biology of Citrus tristeza virus defective RNAs. O. BATUMAN, X. Che, Y. Moskowits, O. Cohen, M. Mawassi, and M. Bar-Joseph. The S. Tolkowsky Laboratory, ARO, The Volcani Center, Bet Dagan 50250, Israel. Phytopathology 94:S131. Publication no. P-2004-0076-SSA.

Isolates of Citrus tristeza virus (CTV) show a uniform genomic organization, despite the considerable variation in their biological properties and genomic composition. Defective (d) RNAs of CTV belong to at least six classes. Class 1, dRNAs with 5′ and 3′ sequences of different lengths, and junction sites non-flanked or flanked by direct repeats of 4-5 nts. Class 2, dRNAs with 3′ moieties corresponding to the full-length ORF11 sgRNA. Class 3, large ca. 12 kb molecules designated LdRNAs1 with 5′ termini corresponding to the 5′ sgRNA of ORF1a+1b (LaMT) and 3′ termini of different sizes. These characteristics are close to the RNA1 genomes of Criniviruses. LdRNAs1 are self-replicating and infectious when transmitted mechanically to citrus plants and *Nicotiana benthamiana* protoplasts. The finding of intact sgRNAs as termini in two classes of dRNAs, suggested that both 5′ and 3′ sgRNAs could be directly involved in RNA recombination. Class 4, large ca. 9.0 kb molecules designated LdRNAs2, which retained all or most of the 3′ ORFs, are hence analogous to RNA 2 in Crinivirus genomes. Class 5, dRNA molecules of 1.7 to 5.1 kb, comprised of the two termini and a non-contiguous internal sequence of >100 nts from ORF2, indicating double recombination, were designated as DR-dRNAs. Interestingly, class 4 dRNAs and DR-dRNAs showed identical size of 5′ portions (948 nts). Class 6, dRNAs with variable size of 5′ and 3′ termini joined by short (14-16 nts) sequences with no homology to the CTV genome. Possible roles of dRNAs in Closteroviridae evolution and CTV biology will be discussed.

An overview of the epidemiology of Citrus tristeza virus. T. R. GOTTWALD. USDA-ARS-USHRL, Ft. Pierce, FL. Phytopathology 94:S131. Publication no. P-2004-0077-SSA.

Epidemics of mild and decline-inducing isolates of Citrus tristeza virus (CTV) were assessed in Florida, Costa Rica, The Dominican Republic, Puerto Rico, and Spain, where the predominant vector for the pathosystem was either *Aphis gossypii* or *Toxoptera citricida*. The vector population composition affected the spatial pattern and rate of CTV increase. In the *A. gossypii* CTV pathosystem, virus incidence reached an asymptote after 8-14 years and was spread through a combination of random transmission originating from inoculum outside the plot and local transmission from within-plot sources operating over short distances. In contrast, virus incidence of the *T. citricida* CTV pathosystem required only 2-4 years to reach an asymptote, and spread was accounted for by short-range transmission within a local area of influence. Long distance spread of CTV was documented by *A. gossypii* over many kilometers, and spread by *T. citricida* was recorded up to 4.0 km. Virus increase was most rapid in orange, slower in grapefruit, and even slower in lemon plantings, although there was little difference in the resulting spatial pattern. Vector control had little effect on CTV increase and spread. Insecticides suppressed aphid infestation of citrus, but aphid feeding occurred on insecticide-treated trees prior to aphid death, and was sufficient for CTV acquisition and transmission.

Transmissibility and genotype analysis of Central California isolates of *Citrus tristeza closterovirus*. R. K. YOKOMI. USDA, ARS, EIDP, Parlier, CA 93648. Phytopathology 94:S131. Publication no. P-2004-0078-SSA.

To ascertain the threat posed by *Citrus tristeza closterovirus* (CTV) in Central California, spread of CTV was monitored from 1997 to 2003 in selected CTV "hot spots". In Tulare Co. where CTV eradication stopped in 1996, incidences up to 42% were found with annual spread rates from 1.6 to 3.6%. In Kern Co. where eradication continues, incidence in a plot in a 4-year-old orchard went from 0 to 5% over a 3-year period before infected trees were eradicated. Fifty field isolates were collected and vector transmissibility examined using standardized conditions and 5 to 10 *Aphis gossypii* per receptor plant. The isolates fell into two broad transmission categories: low (zero to 8%) and moderately high (16 to 64%) which remained consistent for an isolate (e.g. always high or always low). Nearly all isolates characterized had the same genotype as the T30 mild strain from Florida. A few isolates had a non-standard genotype. Aphid transmission did not result in genotype pattern changes, although some sequence variations did occur with some aphid transmitted sub-isolates. These results show that significant reservoirs of CTV now exist, some isolates are highly aphid transmissible, and the genotype of most local isolates is associated with a mild CTV strain. Efforts are now underway to sequence isolates of high and low transmissibility in an attempt to determine which gene(s) control vector transmission.

***Citrus tristeza virus*: Evolution in a host-limited pathosystem.** M. E. HILF. USDA-ARS-USHRL, 2001 S. Rock Road, Fort Pierce, FL. Phytopathology 94:S131. Publication no. P-2004-0079-SSA.

Citrus tristeza virus (CTV) is a major viral pathogen of citrus, with a natural and experimental host range confined to members of the family *Rutaceae*, including the economically important genus *Citrus*. Experimental and agricultural paths of distribution of CTV are transmission by aphids and graft propagation of infected plant material. Analysis of the genomic sequences of five CTV strains suggested that three strains evolved wholly from a common ancestor while two strains appeared to be recombinants between contemporary strains and two different species of closterovirus. The genomic positions of suspected points of recombination and the conserved nucleotide sequence of the 3′ terminal genes suggests that these genes are maintained as a cluster or module, possibly by providing cell-to-cell and long distance movement functions in *Citrus* hosts. Recombination thus may provide a method of increasing genetic variability in this host-limited virus, creating new strains of CTV by linking genes competent for movement in *Citrus* with genes from other viruses competent for replication but not movement in *Citrus*.

Citrus tristeza virus genome encodes three distinct suppressors of RNA silencing. R. Lu (1), A. Folimonov (2), W.-X. Li (1), M. Shintaku (1), B. W. Falk (3), W. O. Dawson (2), and S. W. DING (1). (1) Dept. of Plant Pathol., Univ. of California, Riverside; (2) Dept. of Plant Pathol., Univ. of Florida, CREC, Lake Alfred; (3) Dept. of Plant Pathol., Univ. of California, Davis. Phytopathology 94:S131. Publication no. P-2004-0080-SSA.

Citrus tristeza virus (CTV) is the most destructive virus of the citrus industry worldwide. The large plus-stranded RNA genome (19.3 kb) of CTV encodes 12 open reading frames in total, ten of which are expressed through a series of 3′-coterminal subgenomic RNAs (sgRNAs) although the 5′-proximal ORFs essential for viral replication are directly translated from the genomic RNA. In this study, we analyzed the strategies by which CTV employs to overcome the potent RNA silencing antiviral defense. Two complementary approaches were used to assay for silencing suppression by CTV proteins. The transient expression delivered by agro-infiltration in Nicotiana benthamiana is ideal for identifying suppressors of local silencing. However, grafting experiments that involve the use of stable transgenic tobacco lines is able to identify suppressors of systemic silencing regardless of whether or not they are also active suppressors of local silencing. Our results show that CTV encodes at least three silencing suppressors, all of which are encoded by the more abundantly transcribed sgRNAs of CTV. p23 suppressed local silencing but exhibited no detectable activity in preventing systemic silencing. Similarly, coat protein (CP) was a suppressor of systemic silencing but inactive in the suppression of local silencing. In contrast, p20 suppressed both local and systemic silencing in both assays. However, although p20 was as effective as CP in the suppression of systemic silencing, it was a much weaker suppressor of local silencing as compared to p23. These findings show that viruses may encode more than one suppressor of the RNA silencing antiviral response and that viral interference of distinct steps in the defense pathway may be necessary to ensure an efficient infection.

Positional cloning of the *Citrus tristeza virus* resistance gene. T. E. MIRKOV (1), Z. N. Yang (1), M. Rai (1), J. Molina (1), X. R. Ye (2), and M. L. Roose (2). (1) Texas A&M University, Weslaco, TX; (2) University of California, Riverside, CA. Phytopathology 94:S132. Publication no. P-2004-0081-SSA.

Citrus tristeza virus (CTV) can be a devastating disease of citrus, causing economic losses by killing trees or reducing fruit size and yield. Aphids transmit the virus making control difficult. All commercially grown *Citrus* is susceptible to CTV, but the level of damage varies with cultivar and viral strain. Development and use of resistant varieties will minimize damage to new plantings. The objective of this project is to use positional cloning methods to isolate a dominant gene (*Ctv*) from the *Poncirus* genome that causes resistance to the virus. The gene will then be transformed into CTV-susceptible *Citrus* cultivars to produce CTV-resistant plants. Initial genetic and physical mapping completed during this project delimited the region of the *Poncirus* genome that must contain *Ctv* to a contig of four overlapping BACs that span 300 kb, and complete sequencing of the four BACs to 8X coverage has been completed. 22 genes were identified in this region. Genetic mapping with new microsatellite markers identified by sequencing of the contig allowed us to further delimit the region that contains *Ctv* to 121 kb that contains only 10 genes. Currently, 3 susceptible *Citrus* cultivars (Rio Red, Ruby Red, and Duncan grapefruit) have been transformed with candidate genes. A total of 24 independent transgenic plants, representing 7 of the 10 candidate genes, have been obtained to date. Progress to evaluate the effects of these transgenes on resistance to CTV will be presented.

Fungal Melanins: Biology and Pathogenesis

Biochemistry and genetics of fungal melanin biosynthesis. M. H. WHEELER (1) and H.-F. Tsai (2). (1) USDA-ARS-SPARC, College Station, TX; (2) NIH-NIAID, Bethesda, MD. Phytopathology 94:S132. Publication no. P-2004-0082-SSA.

The importance of melanins for survival and longevity of fungi and their role as virulence factors in certain fungal diseases of plants and animals are now recognized. However, important research contributions continue to be made in these areas. The biosynthetic pathways for different fungal melanins have been described, but the one best characterized is the 1,8-dihydroxynaphthalene (DHN) melanin pathway that occurs in a large number of imperfect and ascomycetous fungi. This presentation is a discussion of past and present research involving several different types of melanin and melanin pathways. It emphasizes the DHN pathway that synthesizes brown to black melanins and compares the enzymes and precursors of this pathway with those that synthesize the green and bluish conidial pigments in *Aspergillus nidulans* and *A. fumigatus*, respectively. Chemical and genetic techniques that have been used in studying melanin pathways and their enzymes are summarized and evaluated, and the synthesis of phytotoxins and other polyketide metabolites from precursors in the DHN pathway is described. Finally, concerns and needed research in the area of fungal melanins are discussed.

The mechanical value of fungal melanin. N. P. MONEY. Miami University, Oxford, OH. Phytopathology 94:S132. Publication no. P-2004-0083-SSA.

Pigment biosynthesis is recognized as a virulence factor for melanotic fungal pathogens of plants and animals. Plant pathologists and medical mycologists studying these microorganisms have found that the inhibition of melanin synthesis results in diverse and severe disabilities for albino cells, particularly when they are challenged by physical and chemical obstacles presented by their hosts. In this presentation, the way in which pigment synthesis affects the biomechanical characteristics of fungal cells will be examined. The link between appressorial melanization, osmolyte accumulation, turgor elevation, and plant penetration is the classic example of this phenomenon. The evolution of this fascinating mechanism is viewed as an arms race between fungus and plant, in which appressorial physiology has been modified in response to changes in leaf architecture (and vice versa). Appressorial melanin seems to work by reducing the permeability of the cell, allowing it to accumulate high concentrations of osmolytes. Melanin does not seem to have the same effect in other pathogenic fungi. With some lateral thinking, however, the appressorial model may help make sense of the value of melanin in a variety of fungi.

Contribution of melanin to pathogenesis of the human pathogen *Cryptococcus neoformans*. A. CASADEVALL, H. Eisenman, J. Garcia-Rivera, A. Mednick, and J. Nosanchuk. Albert Einstein College of Medicine, Bronx, NY. Phytopathology 94:S132. Publication no. P-2004-0084-SSA.

Human pathogenic fungi with melanotic potential include *Cryptococcus neoformans*, *Histoplasma capsulatum*, *Sporothrix schenckii*, and *Paracoccidioides brasiliensis*. Of these, *C. neoformans* has emerged as a model system to study the role of melanogenesis in mammalian fungal disease. *C. neoformans* has a laccase that synthesizes DOPA-like melanins from exogenous substrates. Melanin is deposited in the cell wall, where it can protect the cell against external insults, and cross-linked melanin is made during mammalian infection. Melanized cells are less susceptible than non-melanized cells to oxygen- and nitrogen-derived oxidants, suggesting a mechanism by which melanin could contribute to virulence. Melanized cells are less susceptible to certain types of antifungal drugs, such as amphotericin B and caspofungin, but the pigment does not protect against azoles. Melanin has strong immunomodulatory properties that can interfere with effective host responses. Melanin appears to function in virulence by reducing the vulnerability of fungal cells to host microbicidal mechanisms and altering the host immune response.

The role of melanin production in Ascochyta blight of chickpea. W. CHEN (1), K. D. Sharma (1), M. H. Wheeler (2), and F. J. Muehlbauer (1). (1) USDA-ARS, Washington State University, Pullman, WA 99164; (2) USDA-ARS, College Station, TX 77840. Phytopathology 94:S132. Publication no. P-2004-0085-SSA.

Ascochyta blight caused by *Ascochyta rabiei* is an important disease of chickpea. Two pathotypes of the pathogen are identified in the US. *A. rabiei* produces melanin in culture and in infected plants. The role of melanin production by *A. rabiei* in pathogenicity was investigated. Albino mutants of *A. rabiei*, in contrast to wild types, lost pathogenicity on chickpea. The mutants were able to convert scytalone, a precursor of the 1, 8-dihydroxalnapathalene (DHN) melanin, into a dark compound similar to melanin. Specific melanin-inhibitors, pyroquilon and tricyclazole, blocked melanin production by wild types, suggesting that *A. rabiei* uses the DHN pathway for melanin synthesis. The same specific melanin-inhibitors when applied to plants significantly reduced disease severity caused by wild type strains. Transcripts of scytalone dehydratase, an intermediate enzyme in the DHN-melanin pathway, were detected in conidia using RT-PCR. Data suggest that melanin biosynthesis is operative during spore germination, and melanin production plays important roles in pathogenesis of *A. rabiei* on chickpea.

Morphological dynamics of *Gaeumannomyces* during pathogenesis of roots (take-all). H. T. WILKINSON, H. M. Fouly, and S. W. Henning. University of Illinois, Urbana, IL. Phytopathology 94:S132. Publication no. P-2004-0086-SSA.

The nature of the association (parasitic, pathogenic, saprophytic) between *Gaeumannomyces* and reported host plants is difficult to discern from literature reports that describe host ranges. Research will be reviewed and new research discussed that indicates that the nature of the association between *Geaumannomyces* and a living root is parasitic, but can change to pathogenic and ultimately terminate as saprophytic. As pathogenesis is unraveled, associated changes in hyphal morphology appear to parallel changes in the nature of the association between the fungus and the root. In particular, diameter, shape and melanin content have been observed to changes as various tissues of the root are invaded and colonized. The various roles of melanin are purported to play in the survival and pathogenesis of *Geaumannomyces* will be reviewed and new research findings discussed.

Fungal melanin as a critical component for appressorial penetration by the rice blast fungus. B. VALENT and G. Valdovinos-Ponce. Department of Plant Pathology, Kansas State University, Manhattan, KS 66503. Phytopathology 94:S132. Publication no. P-2004-0087-SSA.

Appressorial melanin is essential for generating turgor pressure that allows the rice blast fungus to breach the rice leaf surface. Mutational analyses identified 3 unlinked biosynthesis genes that are required for pathogenicity, and a 4th gene with no pathogenicity defect in gene disruption mutants. *ALB1* encodes the polyketide synthase involved in production of 1,3,6,8-tetrahydroxynaphthalene (4HN), and *RSY1* encodes scytalone dehydratase (SD) involved in 2 dehydration steps leading 1,8-dihydroxynaphthalene. Two reductases have been identified and biochemically characterized: *BUF1* encodes 1,3,8-trihydroxynaphthalene reductase (also called *3HNR*) with a 4-fold higher preference for 3HN over 4HN, and *4HNR* encodes a second reductase with a 300-fold preference for 4HN over 3HN. Interestingly, *3HNR* is absolutely required for pathogenicity, but *4HNR* is not required in the

presence of a wild type *3HNR* gene. Commercial rice blast fungicides target the SD and 3HNR enzymes. X-ray structures of SD, 3HNR and 4HNR explain substrate and inhibitor binding properties and allow structure-based design of new fungicides. Such a program has produced picomolar inhibitors and potential new fungicides targeting SD. A transcription factor, *PIG1*, regulates expression of melanin biosynthesis genes in mycelium, but not in the appressorium. Future interest lies in identification of transcription factor(s) regulating appressorial melanization and co-regulated genes.

Host–Microbe Interactions in Woody Plants

Genomic approaches to understanding quantitative inheritance of disease resistance in forest trees. D. B. NEALE (1,2), E. S. Ersoz (1), G. R. Brown (1), A. Morse (3), and J. M. Davis (3). (1) University of California, Davis, CA; (2) Pacific Southwest Research Station, USDA Forest Service, Davis, CA; (3) University of Florida, Gainesville, FL. Phytopathology 94:S133. Publication no. P-2004-0088-SSA.

Genetic resistance to pathogens in forest trees may be controlled by single genes (qualitative resistance) or by many genes (quantitative resistance). We are taking a genomics approach toward discovery of quantitative resistance genes. The approach is called association genetics and has been used successfully to dissect complex diseases in humans. Association genetics identifies single nucleotide polymorphisms (SNPs) in linkage disequilibrium with allelic variants functionally responsible for quantitative resistance. To begin, SNPs are discovered in candidate genes by direct DNA sequencing and nucleotide diversity and linkage disequilibrium can be estimated. Next, disease phenotypes and SNP genotypes in candidate genes are determined in the full association population. Statistical tests are performed to test for differences in phenotypic means among SNP genotypic classes. Significant tests are evidence that individual candidate genes are in part responsible for quantitative resistance. We will report on our progress to date to identify candidate gene SNPs associated with quantitative resistance to two important pathogens on loblolly pine (*Pinus taeda* L.); pitch canker (*Fusarium circinatum*) and fusiform rust (*Cronartium quercuum* f. sp. *fusiforme*).

Genetic architecture of loblolly pine interactions with contrasting pathogens. J. M. DAVIS (1), A. M. Morse (1), D. A. Huber (1), C. D. Nelson (2), and S. F. Covert (3). (1) University of Florida, Gainesville, FL; (2) USDA Forest Service, Saucier, MS; (3) University of Georgia, Athens, GA. Phytopathology 94:S133. Publication no. P-2004-0089-SSA.

Loblolly pine (*Pinus taeda* L.) is an ecologically important species, with a native range that spans 14 states where it is the dominant tree species on over 10 million ha of native forest. It is also economically important, since over 1 billion seedlings per year of this species are planted in the U.S. alone. We determined disease phenotypes for clonally propagated loblolly pine trees that were inoculated with either of two distinct, endemic pathogens. *Fusarium circinatum* is a facultative necrotrophic fungus that incites pitch canker disease. *Cronartium quercuum* f. sp. *fusiforme* is an obligate biotrophic fungus that incites fusiform rust disease. Quantitative genetic analysis revealed contrasts among disease traits, and pointed out informative subsets of loblolly pine genotypes useful for identifying disease-related candidate genes. This work is a component of a larger effort to identify trait loci that underlie disease resistance traits.

Anatomy, histochemistry, and cytochemistry of host-pathogen interactions in conifers. V. R. FRANCESCHI. Washington State University, Pullman, WA. Phytopathology 94:S133. Publication no. P-2004-0090-SSA.

Anatomical studies of pathogen-infected tissues provide a basic understanding of disease progression and how tissues react to invasive organisms. Histochemical techniques can further elucidate chemical and biochemical reactions. An overview of techniques is provided to demonstrate their value in understanding host-pathogen interactions and phenomena such as acquired resistance. Anatomical investigations can be applied to samples taken over a time series or exposed to various pathogens or treatment. Cryosections quickly provide histological data, particularly when combined with fluorescent imaging or SEM. Resin embedded sections provide excellent cellular and ultrastructural details using light microscopy and TEM. These sections can be probed with histochemical stains to determine how assimilates and cellular features change in response to a pathogen. Finally, biochemical reactions can be assessed using immunohistochemical and in situ hybridization techniques. As will be shown, these microscopy techniques can be combined with chemical and biochemical analyses to elucidate complex pathways and reactions at high temporal, spatial and chemical resolution.

Fine anatomy and chemical characterization of compartmentalization processes in response to Dutch elm disease and Scleroderris canker. D. RIOUX. Canadian Forest Service, Sainte-Foy (Québec), Canada. Phytopathology 94:S133. Publication no. P-2004-0091-SSA.

Compartmentalization deals mainly with anatomical changes such as barrier zones and wall 3 reaction zones that confine damage resulting from injuries or infections. Barrier zones were found to be continuous in the xylem of nonhost trees inoculated with the Dutch elm disease pathogen whereas they were absent or discontinuous in *Ulmus americana*. A nonhost, *Populus balsamifera*, also responded to inoculation by forming a suberized layer between the pith and the invaded xylem that presented similarities with suberized bands described in a herbaceous wilt-infected plant. Lignin and suberin were major wall constituents of most compartmentalizing cells and phenols were frequently detected in their cytoplasm. Numerous gels and tyloses apparently improved the efficiency of the wall 3 and immunocytochemical tests revealed that pectin is secreted during the development of both structures. Lignosuberized boundaries were also found in *Pinus banksiana* and *P. contorta* resistant to the European race of the Scleroderris canker pathogen. Reacting parenchyma cells also contained phenols, particularly catechins and condensed tannins. Ultrastructural details of the cells involved in compartmentalization are presented. Findings suggesting that embolism is the primary trigger of compartmentalization are discussed as is the role of abscisic acid.

Biochemistry of localized and systemic induced defense responses in pine. P. BONELLO. The Ohio State University, Columbus, OH. Phytopathology 94:S133. Publication no. P-2004-0092-SSA.

Many parallels exist between biochemical defense responses in conifers and in herbaceous model plants. In both, the interface between host and pathogen is often characterized by a hypersensitive response, accompanied by the accumulation of soluble and cell wall-bound secondary metabolites and pathogenesis-related proteins. However, a major role in defense is played in conifers by the resin system, which is absent in herbaceous plants. Furthermore, whereas the occurrence of systemic induced resistance in conifers has been established, the signaling systems underlying it are much less characterized in conifers than in herbaceous plants. I will review the current state of knowledge of biochemical defense responses in conifers and their relationship to resistance. Specific reference will be made to the Austrian pine-*Sphaeropsis sapinea* pathosystem, a canker model system that is amenable to manipulation and should lead to a better understanding of the processes underlying local and systemic induced resistance in coniferous trees.

Exploring the transcriptome of the ectomycorrhizal symbiosis. A. KOHLER, M. Peter, A. Jambois, P. E. Courty, S. Duplessis, F. Lapeyrie, and F. Martin. INRA, Champenoux, France. Phytopathology 94:S133. Publication no. P-2004-0093-SSA.

The tree rhizosphere hosts a large community of microbes that compete and interact with each other and with plant roots. Within this cortege of microorganisms, ectomycorrhizal fungi are almost ubiquitous. Mycelium of symbiotic fungi and root tips form a novel composite organ, so-called ectomycorrhiza, which is the site of nutrient transfer between the symbionts. To examine gene-activity changes associated with the development of the symbiosis, we have performed expression profiling using cDNA arrays of poplar, eucalypt and ectomycorrhizal fungi. A marked change in the gene expression in the mycobiont and the host-plants was observed at multiple levels: (a) a general activation of the protein synthesis machinery probably supporting an intense cell division/proliferation, (b) an increased accumulation of transcripts coding for cell surface proteins probably involved in the symbiotic interface formation, and (c) the upregulation of energy metabolism in roots. This suggests a highly dynamic environment in which symbionts are sending and receiving signals, are exposed to high levels of stress conditions and are remodeling their tissues. With multiple gene profiling programmes dealing with ectomycorrhizal associations, we will have in the future an unparalleled opportunity to ask which genetic features are responsible for common/divergent traits involved in this symbiosis. Possible breakthroughts will be in characterisation of common transcriptional and transduction networks, identification of novel surface proteins and new insights into unique metabolic routes critical for mycorrhiza functioning.

Relevance of herbaceous plant models to understanding and manipulating disease resistance in woody plants. A. F. BENT. University of Wisconsin, Madison, WI. Phytopathology 94:S133. Publication no. P-2004-0094-SSA.

Woody perennials have many distinguishing features not present in *Arabidopsis*, barley, *Nicotiana*, tomato and other plant species from which

much has been learned recently about the molecular basis of plant disease resistance. However, many elements of the disease resistance found in herbaceous plants are likely to be relevant to woody plants as well. This presentation will briefly point out highlights (a biased set) of what we have learned in the last decade about disease resistance in herbaceous model species, with an eye toward what is relevant to woody plants. Pre-formed defenses, disease tolerance, induced resistance, host-pathogen compatibility factors, and R gene-mediated disease resistance will each be discussed. Research progress is greatly expedited by a facile study system, and the methodologies and research community features that have been most central in fostering progress will be noted. Some of the more popular paradigms for disease resistance research, breeding and genetic engineering will be critically compared and discussed, with some slightly Luddite conclusions drawn. All of these things will be covered in 25 minutes. Really.

Interactions Between Plant Pathogens and Their Vectors

Interface between the anatomy of the whitefly *B. tabaci* vector and begomoviral capsid determinants involved in transmission. J. M. CICERO and J. K. Brown. Department of Plant Sciences, University of Arizona, Tucson, AZ 85721. Phytopathology 94:S134. Publication no. P-2004-0095-SSA.

The whitefly Bemisia tabaci (Genn.) is the sole vector of the genus, Begomovirus (family, Geminiviridae). The whitefly esophagus descends from the mouthparts to the anterior abdomen. The midgut descends from there, circumvents the abdominal hemocoel, and loops back to the anterior, where it connects to its anterior end, between the two caecae. This adnation is developed into a complex system of membrane baffles at the base of the filter organ, which is thought to function in osmoregulation. The filter organ is located inside the anterior hindgut, and is a short, conical, highly compact conglomerate of primitive tissues, including malpighian tissue, and the esophageal terminus, which burrows through the anterior hindgut epithelium, embedding itself within. The hindgut then extends posterior to the anus. Vector-mediated transmission of a suite of viral coat protein mutants revealed that three key amino acids modulate virion-whitefly interactions in the transmission pathway. Mass-spectrometric analysis of extirpated primary salivary glands identified ~75 proteins, some of which are candidate begomoviral receptors.

Molecular characterization of plant-parasitizing trypanosomes killing coconut and oil palm in Latin America. M. DOLLET. CIRAD, TA 30/G Campus International de Baillarguet, 34398 Montpellier Cedex 5 France. Phytopathology 94:S134. Publication no. P-2004-0096-SSA.

Several trypanosomatids (Protozoa, Kinetoplastida) can be found in plants. Some are localized in the phloem and are associated with a wilt, some live in latex vessels and other occur in fruits. Fruits can suffer local damages. But in most cases trypanosomatids do not cause any symptom in latex plant. All have similar morphology, and ultrastructure and are put in an arbitrary genus *Phytomonas*. Insects – Hemiptera of the families Lygaeidae, Coreidae, Pentatomidae- propagate these organisms. Under this apparent uniformity, a large diversity and variabilty is found when using sequences analyses of different target genes. The study of 5S rRNA gene shows that if phloem restricted trypanosomatids form one unique defined group – transmitted exclusively by pentatomid bugs - fruits and latex isolates - are not always distinct. Alignment of sequences of the ribosomal operon including the two Internal Transcribed Spacer (ITS) and analyses of microsatellites included in the ITS confirm the diversity. Inside one well defined group as the phloem one, the SL RNA gene can distinguish two robust subgroups. All molecular informations obtained so far match biological and epidemiological data. Correspondances to taxomic levels are under study.

Mapping of domains of the Beet curly top virus capsid protein involved in viral movement, virion formation, and leafhopper transmission. R. L. GILBERTSON, L.-F. Chen, and M. J. Soto. Department of Plant Pathology, University of California-Davis, 1 Shields Ave, Davis, CA 95616. Phytopathology 94:S134. Publication no. P-2004-0097-SSA.

Plant viruses in the Genus Curtovirus (Family, Geminiviridae) are transmitted by the beet leafhopper and cause curly top disease. A full-length clone of Beet mild curly top virus (BMCTV) was obtained from pepper plants from New Mexico. A leafhopper-protoplast feeding assay was used to establish that cloned BMCTV was leafhopper-transmissible and infectious. The role of the capsid protein (CP) in BMCTV pathogenesis was investigated with a frameshift mutant and alanine scanning mutants. All mutants replicated in tobacco protoplasts at levels comparable to wild-type BMCTV. Most N-terminal mutants were infectious in N. benthamiana, formed virions, and were leafhopper-transmissible. In contrast, the frameshift mutant and most C-terminal mutants were not infectious in N. benthamiana, did not form virions, and were not vector-transmitted. One N-terminal mutant CP49-51 was infectious but did not form virions, suggesting a non-virion form moves long distance in plants. The N-terminal mutant CP25-28 was infectious and formed virions, but was not vector-transmissible.

Cellular and molecular regulation of luteovirus transmission by aphid vectors. F. E. GILDOW (1), D. Cox-Foster (1), and S. M. Gray (2). (1) Penn State University, State College, PA; (2) USDA-ARS, Cornell University, Ithaca, NY. Phytopathology 94:S134. Publication no. P-2004-0098-SSA.

Luteoviruses replicate only in phloem cells of infected plants and are transmitted by aphids in a vector-specific circulative manner. Although luteoviruses are transported through aphid cells, the viruses are not known to replicate in aphids. Ultrastructural studies identified the mechanisms of luteovirus transport through aphid gut and salivary tissues and indicated that vector-specificity is regulated both at cell membranes and basal lamina. Luteovirus-binding proteins specific to aphid species have been identified. The similarity of virus-binding proteins in aphids transmitting the same luteoviruses suggests a conservation of receptors regulating cell recognition of viruses. Independent genetic studies of vector and nonvector clones of aphids indicate that transmission competency is controlled by different alleles. Current work is directed at identifying genetic components defining vector competence of aphids and characterizing gene products involved in virus recognition and transport in aphids.

Rhabdovirus host range: A bug's view. S. A. Hogenhout (1), M. G. REDINBAUGH (2), and E. D. Ammar (1,3). (1) Department of Entomology; (2) USDA-ARS; (3) Molecular and Cellular Imaging Center, Ohio State University, Wooster, OH. Phytopathology 94:S134. Publication no. P-2004-0099-SSA.

Rhabdoviruses include human, animal and plant pathogens, and most are insect-transmitted. We use Maize fine streak virus (MFSV) and *Maize mosaic virus* (MMV) to study what determines rhabdovirus host range. Although MFSV and MMV both infect maize, they are vectored by insect species from different families: the leafhopper *Graminella nigrifrons* transmits MFSV and the planthopper *Peregrinus maidis* transmits MMV. The genome sequences of MFSV and MMV were determined, and these provide clues about viral determinants of host range and vector specificity. Interestingly, RT-PCR with MFSV-specific primers on insects fed on MFSV-infected maize showed that MFSV accumulated in several insects that did not vector the virus. We expect to find similar results for MMV. Confocal microscopy of immunolabeled insects showed MMV in most organs and tissues of *P. maidis*, but the virus appeared to be targeted primarily to nerve cells. We hypothesize that, similarly to *Rabies virus* and other rhabdoviruses, MFSV and MMV may spread primarily via the nervous system of their insect vectors and hosts.

Tospovirus-vector relations that drives the spread of disease. A. KRITZMAN (1), B. Raccah (1), D. Peters (2), and A. Gera (1). (1) Department of Virology, Agricultural Research Organization, The Volcani Center, Bet Dagan 50250, Israel; (2) Department of Virology, University of Wageningen, Wageningen, The Netherlands. Phytopathology 94:S134. Publication no. P-2004-0100-SSA.

Tospoviruses (family Bunyaviridae) cause economically important losses in a wide range of vegetable and ornamental crops worldwide. Since the 1980s, their economic importance is rising. New species and isolates belonging to this genus and new host plants are constantly being reported in various crops. Tospoviruses are transmitted by several species of thrips (Thysanoptera: Thripidae), in a persistent and propagative manner. The relations between Tospoviruses and their vectors are highly specific. Different thrips populations of the same species vary in their efficiency to transmit the Tospoviruses. Viral as well as insect factors are involved in the transmission. The virus infects the thrips and replicates in midgut and salivary gland tissues. The thrips anatomy at the different developmental stages and the route of the virus in the thrips body from its acquisition to transmission will be described. Several anatomical and cytological factors that may affect acquisition and inoculation ability will be evaluated. The role of viral components in infection and transmission processes will be discussed.

Rice: A Model Crop that Has Pushed the Frontiers of Plant Pathology and Our Understanding of Host/Pathogen Interactions

***Xanthomonas oryzae* pv. *oryzae* rax genes required for AvrXa21 activity.** S. W. Lee, F. Goes da Silva, Y. Shen, C. Dardick, S. Burdman, R. Yadav, P. Sharma, and P. C. RONALD. Dept. Plant Pathology, UC Davis, Davis, CA 95616. Phytopathology 94:S135. Publication no. P-2004-0101-SSA.

Components of innate immune systems in both plants and animals share many conserved features. Most notably, they sense the presence of pathogen-associated molecules (PAMs), which represent conserved molecular structures, and avirulence (Avr) factors that are strain specific molecules produced by phytopathogens. Little is known about how plant hosts sense and respond to PAMs or Avr factors at the cell surface. One of the best-characterized examples is the rice Xa21 receptor kinase that mediates recognition of *Xanthomonas oryzae* pv. *oryzae* (Xoo) strains expressing AvrXa21 activity. We have identified eight Xoo genes, falling into three classes, which are required for AvrXa21 activity. raxA, raxB and raxC encode proteins with similarity to components of bacterial type I secretion systems. The raxQ and raxP encoded proteins function in concert to produce phosphoadenosine phosphosulfate (PAPS), an active form of sulfate. The raxSt encoded protein shows similarity with mammalian and bacterial sulfotransferases that use PAPs as the sulfuryl donor. Finally, two genes, RaxH and raxR, encode proteins with similarity to two-components regulatory systems and regulate raxSt expression. Based on our results, we hypothesize that upon sensing of the plant environment, the AvrXa21 molecule is sulfated and then secreted by the RaxABC Type I secretion system making it available for race specific interactions with the rice receptor kinase XA21.

A genomics approach to pathogenicity: Saturation insertional mutagenesis in *Magnaporthe grisea*. S. L. Tucker (1), M. Figueroa (1), N. Galadima (1), C. Soderlund (1), Y. Meng (2), M. L. Farman (2), L. Li (3), J.-R. Xu (3), N. Donofrio (4), T. K. Mitchell (4), R. A. Dean (4), and M. J. ORBACH (1). (1) University of Arizona, Tucson, AZ; (2) University of Kentucky, Lexington, KY; (3) Purdue University, West Lafayette, IN; (4) North Carolina State University, Raleigh, NC. Phytopathology 94:S135. Publication no. P-2004-0102-SSA.

Magnaporthe grisea represents a model organism for the study of fungal pathogenicity and growth. As one part of an NSF-funded project taking a functional genomics approach to understand early events in the recognition and responses between *M. grisea* and rice, we are creating a collection of 50,000 DNA insertion lines in *M. grisea* strain 70-15. Our goal is to disrupt all genes encoded in the *M. grisea* genome in order to determine which genes are important for pathogenicity. To maximize coverage of the genome with insertions, different transformation constructs and transformation methods are being used. DNA was introduced into *M. grisea* by both standard protoplast, and *Agrobacterium tumefaciens*-mediated transformation methods. All 50,000 insertion lines are being analyzed for defects in pathogenicity, metabolism and alterations in morphology. Such a project necessitates development of high throughput methods for the generation and screening of putative transformants. In addition, a database has been created for recording all of the transformant data and making it accessible to the public. Over 45,000 strains have been created and the phenotypes of more than half have been analyzed. This talk will focus on analysis of the pathogenicity mutants, along with advances in techniques for the genetic manipulation of this fungus.

Function, evolution and defense signaling pathway of the Pi2/Pi9 blast resistance gene cluster in rice. B. Zhou (1), S.-H. Qu (1), G.-F. Liu (1), M. Dolan (2), H. Sakaii (2), M. Bellizzi (1), B. Han (3), and G.-L. WANG (1). (1) Department of Plant Pathology, The Ohio State University, OH, USA; (2) DuPont Crop Genetics, Delaware Technology Park 100/200, DE, USA; (3) National Center for Gene Research, Chinese Academy of Sciences, Shanghai, China. Phytopathology 94:S135. Publication no. P-2004-0103-SSA.

Rice blast, caused by Magnaporthe grisea, is one of the devastating diseases in rice. To understand the molecular basis underlying the broad-spectrum resistance to rice blast, we initiated the cloning of two broad-spectrum resistance genes, Pi9 and Pi2. Pi9 was introgressed from the wild rice Oryza minuta and Pi2 was introgressed from the breeding line 5137. High-resolution mapping located these two genes in the same region on the rice chromosome 6. Sequence analysis of the Pi9 genomic region identified six candidate resistance genes (Nbs1-Pi9 to Nbs-Pi9) with a nucleotide-binding site (NBS) and leucine rich repeats (LRRs). Only the transgenic lines transformed with Nbs2-Pi9 showed high level of resistance to rice blast. Interestingly, when a 200 kb fragment was sequenced in the Pi2 region, a similar gene cluster encompassing nine NBS-LRRs candidate resistance genes was identified. Complementation confirmed that the Pi9 allelic gene Nbs-2-Pi2 is Pi2. To understand the evolution of the resistance gene cluster, the Pi2-allelic region in five additional susceptible cultivars has been sequenced. Sequence analysis indicated that duplication, insertion and deletion play an important role on the evolution of the Pi2/Pi9 cluster. Microarray analysis of the Pi2 and Pi9 plants after rice blast infection revealed many novel defense genes associated with the broad-spectrum resistance.

Approaches to achieve durable disease resistance in rice. J. E. LEACH (1), B. Liu (2), P. Manosalva (1), S. Lee (1), J. Wu (2), C. Vera Cruz (2), and H. Leung (2). (1) Kansas State University, Manhattan, KS USA; (2) International Rice Research Institute, Manila, Philippines. Phytopathology 94:S135. Publication no. P-2004-0104-SSA.

Durable resistance to pests and pathogens is a long-sought goal of crop protection programs worldwide because it is considered cost-effective, environmentally sound, and promotes conservation of limited genetic resources. However, realizing durable resistance, which is controlled by both qualitative and quantitative traits, has been limited by the lack of understanding of its molecular basis. While progress with qualitative resistance has been significant, much remains to be discovered about the genes that collectively confer quantitative resistance. In recent years, many rice genes that are candidates for participation in quantitative resistance were cloned and the resources (genetic and mutant stocks and genome microarrays) to critically evaluate their roles in quantitative resistance are becoming available. We used these resources to establish the association of candidate defense genes with quantitative traits for resistance to rice blast, bacterial blight, sheath blight, and brown planthopper. Accumulation of these candidate genes into rice resulted in high levels of disease resistance in farmers' fields. This study provides the basis for analysis of the functions of genes in conferring quantitative resistance and demonstrates how the individual and combined effects of these genes impact durable resistance. Furthermore, it demonstrates that candidate gene selection offers exciting opportunities to solve intractable problems in crop improvement using available genomics information.

Comparing transgenic and conventional resistance to rice hoja blanca virus. L. CALVERT, I. Lozano, Z. Lentini, A. Garavito, N. Villareal, L. Fory, R. Meneses, and M. Triana. CIAT A.A. 6713, Cali, Colombia. Phytopathology 94:S135. Publication no. P-2004-0105-SSA.

Progress has been made in developing rice with resistance to rice hoja blanca virus (RHBV). The virus propagates in the planthopper vector *Tagosodes orizicolus*. Conventional breeding has led to the development of varieties with better resistance to hoja blanca disease than the parental source of resistance Colombia 1. Also RHBV resistant transgenic rice lines have been developed. The host/vector studies indicate that resistance to both the virus and the vector are important for field resistance to RHBV. This plant resistance can be overcome by using high disease pressure with highly aggressive vector colonies, which are adapted to the virus. Gene silencing appears to be one mechanism of resistance to the virus in the transgenic lines. For the conventionally breed resistance, the studies indicate that two or three genes are involved and molecular markers to one of these genes have been identified on chromosome 4. Some of the transgenic rice lines develop a hypersensitive reaction upon infection by RHBV. Studies are underway to determine if the gene silencing is signaling the hypersensitive pathway or if this is an independent mechanism of resistance.

Suppression of Host Defense Responses by Pathogens

Molecular basis of bacterial disease resistance in *Arabidopsis thaliana*. B. J. STASKAWICZ, D. Dalhbeck, P. Coppinger, B. Day, S. Chisholm, G. Coaker, and M. Briggs. Department of Plant and Microbial Biology, University of California, Berkeley, CA 94720. Phytopathology 94:S135. Publication no. P-2004-0106-SSA.

Arabidopsis thaliana has been developed as a model system to elucidate the molecular basis of bacterial disease resistance to infection by *Pseudomonas syringae* pv. *tomato* (Pst) strain DC 3000. Pst DC3000 employs the Type Three Secretion System (TTSS) to directly deliver bacterial effector proteins to the plant host cell. Specifically, our laboratory has studied the molecular events associated with the activation and expression of the RPS2 disease resistance signaling pathway. Evidence will be presented to demonstrate that the RIN4 protein is a negative regulator of RPS2 activity and that the RIN4 protein is a target for the AvrRpt2 cysteine protease. Data will also be

presented that suggests that the degradation of RIN4 by AvrRpt2 activates the RPS2 resistance signaling pathway. Finally, a molecular model will be presented that integrates our current knowledge of the molecular events controlling the RPS2 disease resistance signaling pathway.

***Pseudomonas syringae* type III effectors – suppressors of plant innate immunity.** J. R. ALFANO. University of Nebraska, Lincoln, NE. Phytopathology 94:S136. Publication no. P-2004-0107-SSA.

The *Pseudomonas syringae* pv. *tomato* DC3000 type III secretion system (TTSS) and the effector proteins it delivers into plant cells are required for bacterial pathogenicity on plants and elicitation of the hypersensitive response (HR), a programmed cell death (PCD) that occurs on resistant plants. We have used several screens to determine whether type III effector proteins suppress basal defenses (induced by general elicitors) and/or host specific defenses (induced by Avr proteins). For example, cosmid pHIR11 encodes a functional TTSS and enables nonpathogens to elicit an HR dependent upon the TTSS and Avr protein HopPsyA. We used pHIR11 to determine that the DC3000 effectors HopPtoE, AvrPphE$_{Pto}$, AvrPpiB1$_{Pto}$, AvrPtoB, and HopPtoF could suppress a HopPsyA-dependent HR on tobacco and Arabidopsis. DC3000 effector mutants elicited an enhanced HR consistent with these mutants lacking an HR suppressor. Several of these suppressors inhibited the expression of the tobacco PR1a defense-related gene. Another effector, HopPtoD2, possessed protein tyrosine phosphatase activity and suppressed the HR and an HR-like response initiative by a MAP kinase pathway in tobacco suggesting that HopPtoD2 acts as a suppressor by modifying a MAP kinase pathway. Recently, we have shown that several effectors also suppress callose deposition induced by a flagellin-triggered Fls2-dependent pathway suggesting that, in addition to suppressing the HR and PR1a expression, these suppressors also inhibit basal nonhost resistance in plants. The high proportion of effectors that suppress plant defense suggests that suppressing plant immunity is one of the primary roles for DC3000 effectors and a central requirement for P. syringae pathogenesis.

Fungal effector gene function in pathogenicity and host specificity. B. VALENT, P. Kankanala, M. Dalby, R. Berruyer, S. Poussier, G. Valdovinos-Ponce, and G. Mosquera. Department of Plant Pathology, Kansas State University, Manhattan, KS 66503. Phytopathology 94:S136. Publication no. P-2004-0108-SSA.

Our goal is to understand the dual roles for fungal effector molecules in promoting biotrophic invasion of plant cells and in triggering R-gene mediated resistance. The best characterized fungal effector candidate is the avirulence gene *AVR-Pita* from the hemibiotrophic rice blast fungus. AVR-Pita appears to induce resistance by direct interaction with the Pi-ta resistance protein inside living plant cells. AVR-Pita is a zinc metalloprotease, and we are investigating its general substrate specificity including whether Pi-ta itself is a substrate. We are using fluorescence live cell microscopy and plasmolysis assays to characterize the initial biotrophic stage, and we have initiated studies to demonstrate AVR-Pita secretion into living plant cells. *AVR-Pita* promoter analysis will identify cis-elements responsible for its infection-specific expression. Identification of critical sequence motifs for *AVR-Pita* function will lead to strategies for recognizing additional effector genes in the *M. grisea* genome sequence. In a second strategy to identify fungal effectors, we are pursuing Laser Capture Microdissection (LCM) to selectively purify rice cells containing developing biotrophic invasive hyphae for RNA extraction and microarray analysis. LCM purification of invaded rice epidermal cells 24 to 36 hours after inoculation with fungus, at the time when susceptibility and resistance is decided, will highly enrich for mRNAs that are critical for the infection outcome, including fungal effector gene candidates.

Identification and characterization of glucanase inhibitor proteins: Uncovering the molecular arms race in the plant cell wall. J. K. C. ROSE. Department of Plant Biology, Cornell University, Ithaca, NY. Phytopathology 94:S136. Publication no. P-2004-0109-SSA.

The plant cell wall represents the interface with microbial pathogens and so it is not surprising that many wall-localized mechanisms for surveillance, attack and defense have been identified. We have recently discovered a new class of proteins, termed glucanase inhibitor proteins (GIPs) that are secreted by species of *Phytophthora* into the plant wall during pathogenesis. GIPs bind and inhibit the activity of plant extracellular endo-beta-1,3-glucanases (EGases), thus blocking the release of glucan elicitors. Several GIPs were first identified in *P. sojae* and they appear to show a high specificity for particular EGase isoforms since GIP1 binds specifically to the EGaseA isoform from soybean, but not to another isoform, EGaseB. Despite their potential importance as suppressors of EGase-mediated defense responses, the molecular basis of GIP action and specificity are not well understood. We have recently identified a 4-member GIP family from *P. infestans*, and detected GIP isozymes both in culture and *in vivo* into the apoplast of *P. infestans*-infected tomato leaves using Western analysis. Molecular modeling has been used to predict putative docking sites on the surfaces of EGases and GIPs that may be involved in the high affinity binding between these proteins and the results suggest that positive selection has driven the co-evolution of these protein families. This further suggests the existence of a molecular arms race between GIPs and EGases.

Extracellular protease inhibitors of *Phytophthora infestans* determine a novel counterdefense mechanism. M. TIAN, N. Champouret, and S. Kamoun. Department of Plant Pathology, The Ohio State University, Wooster, OH. Phytopathology 94:S136. Publication no. P-2004-0110-SSA.

The oomycete *Phytophthora infestans* causes late blight, a ravaging disease of potato and tomato. *P. infestans* is a hemibiotrophic pathogen that requires living host cells to establish a successful infection. Suppression of host defenses by *P. infestans* is thought to be a key pathogenicity mechanism, but remains poorly understood. We used data mining of *P. infestans* sequence databases to identify 18 extracellular protease inhibitor genes, belonging to two major structural classes: (i) Kazal-like serine protease inhibitors (EPI1 to EPI14) and (ii) cystatin-like cysteine protease inhibitors (EPIC1 to EPIC4). Eight EPIs and EPICs were expressed in *Escherichia coli* and affinity purified as fusion proteins with the epitope tag FLAG. Recombinant EPI1 specifically inhibited subtilisin A among major serine proteases, and inhibited and interacted with the pathogenesis-related P69B subtilisin-like serine protease of tomato. Interestingly, EPIC1 and EPIC2 were degraded by P69B but EPI1 protected both proteins from degradation. Co-immunoprecipitation experiments revealed that EPIC2 interacts with a novel extracellular cysteine protease of tomato. Overall, our results suggest that complex cascades of inhibition of host proteases occur in the plant apoplast during infection. Both Kazal-like and cystatin-like inhibitors are widespread in the oomycetes, but are absent in other microbial plant pathogens. Inhibition of host proteases by *P. infestans* protease inhibitors is proposed to be a novel mechanism of pathogen suppression of plant defenses.

Plant Disease Management

Organic Foods – From Production to Market

Organic farming and plant disease research by the University of California, 1987-2004. J. C. BROOME. University of California, SAREP, Davis, CA. Phytopathology 94:S136. Publication no. P-2004-0111-SSA.

The University of California Sustainable Agriculture Research and Education Program (SAREP), formed in 1986, has funded over $8 million worth of projects. The projects are included in a database along with an assessment of organic relevance, publications and other outcomes. Twenty-nine multiple year projects were determined to be directly relevant to organic farming, totaled almost $2 million, and included two long term farming systems comparison studies based at UC Davis with plant pathologists and nematologists (SAFS; LTRAS). There have been 63 projects that totaled $4.3 million deemed indirectly relevant to organic farming. Recently, SAREP has obtained additional funding to support an Organic Initiative within UC Cooperative Extension totaling $753,298. A UC Organic Farming Research Workgroup was formed in 2000 and is comprised of 97 individuals of whom 13 indicate plant disease as being a focus of their work. These individuals come from UC, USDA-ARS, and government agencies within California. A UC wide survey will be conducted in April 2004 to assess organic farming research activity and will be analyzed for overlap of plant disease and organic farming research, and these results will also be presented.

Best management practices on organic farms provides opportunity and challenge for applied research plant pathology. D. O'BRIEN. O'Brien Agricultural Consulting, Organic Farming Research Foundation, Santa Cruz, CA. Phytopathology 94:S136. Publication no. P-2004-0112-SSA.

Best Management Organic Practices (BMOP) are replacing input substitution among successful organic farmers. Use of BMOP is a whole-ecosystem (rather than reductionist) approach that acknowledges the dynamic relationships among multiple elements of the farm. Interest and funding for BMOP research is increasing and collaborators in plant pathology are needed. Five

years of reviewing competitive organic research grant proposals and fifteen years of in-field work with many North American organic vegetable farmers suggests that BMOP helps to achieve improved ecological, societal and economic agriculture. Observations on Salinas Valley, U.S.A.-area organic vegetable farms suggest that BMOP improves control of several diseases; for example: downy mildew of lettuce caused by *Bremia lactucae*, and root rot of peppers caused by *Phytophthora* spp. Conversely, BMOP seems to increase damage in a few diseases such as pink root of onion caused by *Phoma terrestris*, and a watermelon wilt possibly caused by *Fusarium solani*.

Farming systems research and extension in organic agriculture: A plant pathology perspective. F. J. LOUWS. North Carolina State University, Raleigh, NC. Phytopathology 94:S137. Publication no. P-2004-0113-SSA.

The science of plant pathology includes the search to acquire basic knowledge of pathogen biology, environmental variables, management of the crop, and the complex interactions that lead to unacceptable economic losses. This necessitates focused research and extension information on specific tactics to manage a distinct pest within a single crop. Organic farmers also seek increasingly complex information that integrates capabilities to maximize benefits associated with complex farming systems, enhanced biodiversity, and beneficial ecological functions of biota, i.e. growing multiple crops over time and space to foster greater biodiversity, multi-pest suppression, and vigorous plant health. The discipline goal to study specific tactics of disease control and the interdisciplinary goal to study large scale farming systems are complimentary and not necessarily mutually exclusive. Likewise, extending information is best appreciated by combining specific and systems-based knowledge. At NCSU, we have implemented a farming systems experiment at the Center for Environmental Farming Systems (sustainable-ag.ncsu.edu/cefs/), conducted on-farm research to address specific issues, developed graduate credit courses in Organic Farming Systems (HortTechnology 10:675-681) and On-Farm Research, and participated in the development of a national Organic Agriculture Consortium that generates new knowledge and centrally extends organic production information (organicaginfo.org). Plant pathologists can offer specific and team-based information to enhance the organic foods industry.

Compost teas: A tool for rhizosphere + phyllosphere agriculture. S. DIVER. ATTRA, National Sustainable Agriculture Information Service, Fayetteville, AR. Phytopathology 94:S137. Publication no. P-2004-0114-SSA.

Compost teas—aqueous extracts of compost for soil and foliar application to gain plant growth promotion and disease suppression in the rhizosphere and phyllosphere—have become a hot topic in organic farming. Practitioners and private industry laboratories have developed sophisticated methods of aerobic brewing, compost tea additives to promote beneficial microbe growth, microbial analysis, and application uses. However, the National Organic Program treats compost teas the same as raw manures in Section 205.203. Compost teas cannot be applied to crops within 90/120 days prior to harvest, thus eliminating compost tea application from the growth life cycle for most crops. An NOSB Compost Tea Task Force was formed to examine human bacterial pathogen risk from compost tea application, especially from manure-based composts, and to develop updated guidelines and recommendations.

Inclusion of crop protection materials on the National Organic Program's national list of allowed substances. R. L. KOENIG. Rosies' Organic Farm, National Organic Standards Board, University of Florida, Gainesville, FL. Phytopathology 94:S137. Publication no. P-2004-0115-SSA.

The National Organic Program Standards require that producers use a planned systems approach to crop protection. This is achieved by utilizing cultural, mechanical, physical, and biological control methods. Non-synthetic biological, botanical, or mineral inputs may be employed to manage diseases; however, these substances are allowed only when the practices described above are insufficient to prevent or control diseases. The Organic Food Production Act (OFPA) requires the Secretary of Agriculture to establish a National List of Allowed Substances which identifies synthetic substances that may be used in organic production. The OFPA authorizes the NOSB to develop and forward to the Secretary of Agriculture a recommended proposed National List, and subsequent proposed amendments to it. To add a substance to the National List, it must be determined that the use of such substances would not be harmful to human health or the environment, is necessary to the production or handling of the agricultural product because of unavailability of wholly natural substitute products, and is consistent with organic farming and handling (7 USC 6517).

Funding opportunities for integrated projects specific to organic agriculture. T. A. BEWICK. USDA/CSREES/PAS, Washington, DC. Phytopathology 94:S137. Publication no. P-2004-0116-SSA.

The Cooperative State Research Education and Extension Service has two authorizations that allow us to fund integrated competitive programs for organic agriculture. The first is the Organic Transitions Program. Grants were first issued in 2001. Since then 15 projects have been funded through 2003. Awards ranged from $498,000 (4 yr) to $93,000 (2 yr). As a result of this program, at least indirectly, there has been a 46% increase in certified organic acres at U.S. land-grant institutions. The second authorization is for the Organic Research and Extension Initiative, which was competed for the first time in 2004. This is a mandatory program funded at $3 million per year for five years.

Sampling for Pathogen Detection to meet Quarantine and Certification Requirements

Strategies of sampling for detection. G. HUGHES (1), L. V. Madden (2), and T. R. Gottwald (3). (1) University of Edinburgh, Edinburgh, UK; (2) OARDC, Ohio State University, Wooster, OH; (3) USDA-ARS, US Horticultural Research Laboratory, Fort Pierce, FL. Phytopathology 94:S137. Publication no. P-2004-0117-SSA.

Strategies of sampling for detection employ the methodology of acceptance sampling. Quarantine inspection of fruits, vegetables and other plant products implies a decision-making process that may result in acceptance or rejection of the whole of a shipment on the basis of a sample. Such sampling schemes can be defined by the sample size and the acceptance number. The latter is the maximum number of defectives allowed in a sample of the appropriate size, for the shipment still to be judged acceptable. In the extreme, the acceptance number may be zero. The formulation of an acceptance sampling scheme for use in quarantine inspection reflects the rates at which we are prepared to wrongly reject shipments that are truly acceptable and to wrongly accept shipments that are truly defective. These rates are not necessarily the same. The principles of acceptance sampling are also applicable in other situations where sampling is used to support a decision-making process relating to plant health, including certification schemes and monitoring in the context of eradication programs.

Looking for viruses in all the right places: New vectors and viruses in small fruit crops. R. R. MARTIN. USDA-ARS-HCRL, Corvallis, OR 97330. Phytopathology 94:S137. Publication no. P-2004-0118-SSA.

During the last 15 years a number of new and reemerging virus diseases have become important constraints in production of small fruit crops in North America. Pollen- and aphid-borne viruses have become important factors in blueberry, raspberry and strawberry production. Concentrated production and planting of susceptible cultivars has led *Raspberry bushy dwarf virus* to reemerge as a serious disease in *Rubus* species worldwide. In strawberry, some aphid-borne viruses have expanded their geographical ranges. The expansion is not due to a change in the range of the vector, but rather a change in virus-vector relations. In strawberry and blackberry, the naturalization of the greenhouse whitefly *Trialeuroides vaporariorum*, and possibly other whiteflies in the southern regions of the USA has resulted in the widespread appearance of several new criniviruses as well as *Beet pseudo-yellows virus*. A complex pattern of plant production and movement also contributes to the increase in virus and phytoplasma diseases in production fields. The detection and impact of these viruses, along with the occurrence of viruses new to the USA will be discussed.

Innovations in detection technology to limit the dissemination of phytopathogens in seed. R. R. WALCOTT. Department of Plant Pathology, The University of Georgia, Athens, GA 30602. Phytopathology 94:S137. Publication no. P-2004-0119-SSA.

Because planting seeds are internationally traded commodities, they represent a potential weak link in US plant biosecurity. Seeds may become infested with plant pathogens, and thus serve as vectors to move pathogens across geopolitical borders. To guard against this, seed health testing is widely employed; however, current seed detection technology lacks the sensitivity, specificity and efficiency to completely exclude seedborne pathogens. The polymerase chain reaction (PCR) is a powerful assay, with the potential to significantly improve seed health testing. However, extraction of PCR-quality pathogen DNA from seeds is difficult because epidemiologically significant pathogen populations can be low. More importantly, seeds contain compounds that inhibit PCR, yielding false negative results. The ability to

detect pathogens can be significantly improved by developing protocols that efficiently yield high quality target DNA from seeds. In recent years, protocols such as enrichment PCR, immunomagnetic separation (IMS)-PCR and magnetic capture hybridization (MCH)-PCR have been applied as seed detection assays. These techniques seek to selectively concentrate target pathogens/DNA while simultaneously eliminating PCR inhibitors. As such, they demonstrate improved efficiency and sensitivity over conventional assays. Additionally, the availability of real-time PCR technology increases the efficiency of PCR and makes the simultaneous detection of multiple seedborne pathogens in seeds. This paper will discuss recent innovations in seed detection technology, with a specific focus on the potential benefits of IMS-PCR and MCH-PCR.

Clean stock programs and virus detection in vegetatively propagated ornamental plants. J. A. DODDS. Dept. Plant Pathology, University of California, Riverside. Phytopathology 94:S138. Publication no. P-2004-0120-SSA.

The ornamental industry encompasses many viruses and plant species. Vegetative propagation from nuclear stock is increasingly common, with larger operations containing hundreds of species and cultivars. It is often difficult to keep well-tested stock separate from stock that could be a source of virus re-introduction. There is an increasing need to ensure that nuclear stocks are free from viruses and propagation stock is tested sufficiently to maintain confidence in low or no virus incidence. Horticulturalists who develop new lines do not always pay attention to viruses, especially those that cause latent or mild infections. Virologists need to establish logical programs for virus testing and certifications with particular emphasis on sampling. In vitro propagules may or may not have the same kind of virus titer and distribution patterns than mature plant parts, and propagules that test negative need to be retested when mature plants are available. The large number of viruses found in the ornamental industry in part reflects the high incidence of multiple infections. The multiple viruses found in Petunia are a case in point. The need for timely dissemination of knowledge when specific viruses are detected in a line is of some importance in an era where the production of nuclear plants, the increase of mother plants and the rooting of cuttings may occur in three different locations, even countries, with the end product destined for none of these places. There will continue to be a need for high quality clean stock programs for management of viruses in ornamental plants.

Ralstonia in geraniums: A case study. M. J. KLOPMEYER. Ball FloraPlant. Phytopathology 94:S138. Publication no. P-2004-0121-SSA.

Over the past two years, *Ralstonia solanacearum* race 3 biovar 2, cause of bacterial wilt, has been introduced into the USA on infected geranium cuttings from off-shore production facilities. The introduction of the pathogen has caused significant losses to growers and suppliers of vegeatively propagated plants. Strict sanitation and sampling are the only means companies have to assure pathogen-free production of ornamentals. Industry's perspective on disease prevention and sampling to meet certification and quarantine requirements will be presented.

Idealism versus reality: When laboratory throughput affects the sampling scheme. L. LEVY (1), D. Kaplan (2), P. Berger (2), and L. Brown (2). (1) USDA APHIS PPQ CPHST, Beltsville, MD; (2) USDA APHIS PPQ CPHST, Raleigh, NC. Phytopathology 94:S138. Publication no. P-2004-0122-SSA.

Sampling schemes and the collection of samples are related to the biology and epidemiology of the organism, as well as parameters of the detection method utilized. Detection limits and protocol complexity (sample preparation and testing) may complicate rapid, large scale, high throughput diagnostics. Techniques that are developed, or validated, often require adaptation to large scale surveys. For regulatory purposes, diagnostic methods must be validated and deployment laboratories should undergo training and proficiency testing to reduce variables that may affect the quality of information collected from large scale surveys. The complexity of sampling and methods must be considered since they will be deployed to laboratories with various skill levels. These points will be discussed in relation to recent events with regulatory pathogens and Select Agents.

The Nature and Application of Biocontrol Microbes II: *Trichoderma* spp.

Overview of new insights into mechanisms and uses of Trichoderma based products. G. E. HARMAN. Cornell University. Phytopathology 94:S138. Publication no. P-2004-0123-SSA.

Trichoderma spp. long have been known to interact with other microorganisms, especially fungi, through antibiosis, mycoparasitism, competition of various types and other mechanisms. More recently, they have been shown also to have strong effects on plants. Induced systemic resistance has been demonstrated with many strains on both monocots and dicots, resulting in control of bacterial, fungal, Oomycete and viral plant pathogens. Control of diseases may be at sites temporally and spatially distant from the site or time of application, especially with strongly rhizosphere competent strains. Root colonizing strains also can enhance plant growth and yield, and, especially, root growth for months after application. This confers resistance to abiotic stresses including inadequate fertilization, drought and soil compaction. However, plant genotype affects these plant growth responses. The fungi also are able to enzymatically degrade a wide range of compounds and are very promising for uses such as pollution bioremediation and for commercial production of chitinases.

Changes in taxonomy, occurrence of the sexual stage and ecology of *Trichoderma* spp. G. J. SAMUELS. USDA/ARS. Phytopathology 94:S138. Publication no. P-2004-0124-SSA.

DNA sequence analysis has had at least three positive effects on taxonomy of *Trichoderma*. All of the approximately 60 described species (including unnamed anamorphs of *Hypocrea*) have been characterized by two or more genes, making identification of species accessible to anybody who can obtain DNA sequences. However, exploration of new niches, such as endophytes, and new geographic locations will result in a substantial increase in the number of species of *Trichoderma*. Molecular phylogenetic analyses have revealed the existence of many more species than have been recognized on the basis of morphology alone but have provided a phylogenetic framework from which predictions of biological activity can be made. DNA sequence data show that *Trichoderma* and *Hypocre* are phylogenetically indistinguishable and most of the named *Trichoderma* species have been linked to teleomorphs in *Hypocrea*. Many of the sectional subdivisions of *Trichoderma* are not monophyletic but their names are useful as morphological descriptors. *Trichoderma* is usually thought of as being a genus of free-living soil fungi but evidence suggests that *Trichoderma* species may be opportunistic, avirulent plant symbionts as well as being parasites of other fungi. *Trichoderma* species are now known to be the dominant endophytes found within trunks of asymptomatic cocoa trees.

Systemic resistance induced by Trichoderma hamatum 382: Interactions between the host, the biocontrol agent, and soil organic matter quality. H. A. J. HOITINK and L. V. Madden. Ohio State University. Phytopathology 94:S138. Publication no. P-2004-0125-SSA.

Trichoderma hamatum 382 (T382) induces ISR throughout treated plants. Most PR protein activation occurs after plants are invaded by a pathogen. Efficacy is directly related to the microbial carrying capacity of the substrate. T382 as some other Trichoderma strains, is more effective in compost-amended than in Sphagnum peat substrates. In nurseries, the severity of Phytophthora leaf blight and of stem dieback of Rhododendron and of Botryosphearia stem dieback on *Myrica pennsylvanica* was reduced significantly in compost-amended media inoculated with T382. However, anthracnose on *Euonymus fortunei* cv Emerald Gold consistently was not affected. Using a strain-specific PCR assay, it was shown that T382 did not spread frequently under commercial conditions from inoculated to control compost-amended media nor to roots or leaves of control plants even though other isolates of *T. hamatum* and other *Trichoderma* spp. were abundant there. Lack of dissemination of ISR-active strains such as T382 may explain why ISR is a rare phenomenon in natural compost-amended substrates.

Understanding the mechanisms employed by *Trichoderma virens* to effect biological control. C. R. HOWELL. USDA/ARS. Phytopathology 94:S138. Publication no. P-2004-0126-SSA.

To make the most effective use of a biological control agent of soil borne diseases, the effects of the microbial and edaphic environment on its activities, and the mechanisms it employs, must be understood. The most obvious features of *Trichoderma virens* are its mycoparasitism of many fungi, including plant pathogens, and its production of the wide spectrum antibiotic, gliotoxin. This has lead many investigators to speculate that either one or both of these mechanisms are responsible for the observed biological control activities of the fungus. However, later research has shown that mutants of *T. virens* deficient for mycoparasitic activity and/or gliotoxin production are still effective biological control agents. The most current research has indicated that two other mecha-

nisms are most likely responsible for observed seedling and root disease control by *T. virens*. One mechanism involves the induction of phytoalexin synthesis in the seedling radicle and plant root through penetration of the outer surfaces by *T. virens* as it grows along the developing root. The second mechanism involves the metabolism, by *T. virens*, of seed emitted inducers of pathogen propagule germination, before these compounds reach the pathogen.

The molecular biology of the interaction between *Trichoderma*, phytopathogenic fungi and plants. M. LORITO and S. L. Woo. University of Naples, Portici, Italy. Phytopathology 94:S139. Publication no. P-2004-0127-SSA.

Trichoderma-based biofungicides are a reality in agriculture, with more than 50 formulations today available as registered products worldwide. Several strategies have been applied to identify the main genes and compounds involved in this complex, three-ways cross-talk between the fungal antagonist, the plant and microbial pathogens. Proteome and genome analysis have greatly enhanced our ability to conduct holistic and genome-based functional studies. We have identified and determined the role of a variety of novel genes and gene-products, including ABC transporters, enzymes and other proteins that produce or act as novel elicitors of Induced Resistance, proteins responsible of a gene-for-gene avirulent interaction between *Trichoderma* and plants, mycoparasitism-related inducers, plant proteins specifically induced by *Trichoderma*, etc. We have transgenically demonstrated the ability of *Trichoderma* to transfer heterologous proteins into plant during root colonization. We have used GFP and other markers to study the interaction in vivo and in situ between *Trichoderma* and the fungal pathogen or the plant.

Professionalism/Service/Outreach

Adapting Teaching Styles and Techniques for a Changing Student Population

How students learn science: What research and experience tell us. R. W. DUNBAR. Stanford University, Stanford, CA. Phytopathology 94:S139. Publication no. P-2004-0128-SSA.

Never before in the history of teaching has there been more practical wisdom and more available research to inform the process by which we create learning experiences for students. Yet unlike our own research, where collaboration and existing literature underpin every step we take, we are more likely in the classroom to teach in isolation, "as we were taught." Initiatives led by the National Science Foundation, National Academy of Sciences, and National Research Council ask us to expand the model of science teaching in colleges and universities—to *teach* science as we *do* science—in an effort to promote deep and meaningful learning. Known contributors to this process are the following research-based factors: 1) Prior knowledge impacts construction of new knowledge; 2) novices need effective organizing schemes; 3) students should be actively engaged in the learning process; 4) feedback should be frequent, timely, and constructive; 5) multiple representations enhance learning for all learners; and 6) understanding the way one learns can improve the learning process. There is no one "best way to teach." We can incorporate best practices in a wide variety of teaching styles, and as we do so, we create learning environments in which science students flourish.

Increasing student diversity: Challenges and opportunities. C. D'ARCY. University of Illinois, Urbana-Champaign. Phytopathology 94:S139. Publication no. P-2004-0129-SSA.

Over the past several decades, student populations at our colleges and universities have become more diverse. Today, we often teach more women, persons of color, international students, older students, students from modest economic backgrounds and, in agriculture, more students from urban/suburban areas than we did in the past. But our students also have changed in other, less visible, ways; the expectations and goals of the "millennials" differ significantly from those of us who are "baby boomers" or "generation Xers". In order to provide today's diverse students equal opportunities to learn, we as teachers also must change. A first step is to become more knowledgeable about our students, which can be done in several ways – through reading about them, communicating with them, or even formally studying them. In the general education plant pathology course at the University of Illinois, we are currently conducting a study to compare our students' preferred learning styles (determined through the Gregorc Style Delineator) with their perceptions of which formats and media are most conducive to their learning. Our goals are to determine (1) the diversity in learning styles among our students, (2) whether we are effectively reaching all learners and (3) what additional instructional formats and media may be required in the course. Like all teachers from past "generations", we must continue to develop as teachers in order to ensure that all students of this generation have the opportunity to succeed as learners.

Utilization of case studies in teaching plant pathology. M. RILEY. Clemson University, Clemson, SC. Phytopathology 94:S139. Publication no. P-2004-0130-SSA.

Increased knowledge retention and increases in the ability of students to relate plant pathology concepts and principles to their future careers can be obtained when using case studies. Different teaching methods have been used to get students actively involved in their own learning as well as interested in the subject. Case studies are a teaching method that has been successfully used in many disciplines especially in training of medical doctors and can also be useful in plant pathology courses. Case studies are real life situations where students are required to do some type of decision making or investigation. There is some problem that the students must be solved. Data and figures relating to the problem can be incorporated into the case. The most successful cases are those where students are actively involved as characters in a case. How case studies can be used in classroom and laboratory situations and the development of cases will be discussed. A case will also be presented using the audience as class members to illustrate activities of instructors when utilizing cases.

Better or just different: New technologies and teaching. D. EASTBURN. University of Illinois, Urbana-Champaign. Phytopathology 94:S139. Publication no. P-2004-0131-SSA.

Teachers have adopted new technologies for teaching many times. Chalkboards, photocopies, and overhead projectors are viewed as standard teaching tools today, but at one time they were cutting edge technologies, and their usefulness for teaching was uncertain. Some technologies are adopted as a convenience for the instructor, while others allow the presentation of information in a whole new way, leading to increased student learning. Teachers today are confronted with an unprecedented array of new information technologies for use in teaching. Instructors are being urged to use presentation software, such as PowerPoint; course management software, such as WebCT; computer based discussion boards and chat rooms; virtual laboratory software; and the internet as a means for enhancing student learning. At the same time, many of today's students have high expectations for the use of information technologies in their courses, but they can be critical of instructors who do not use the technologies effectively. Some of these technologies can be true time savers or effective for enhancing student learning. However, instructors should carefully consider their various advantages and disadvantages.

Plant Pathology in Historical Perspective

Bacteriology and plant pathology in late 19th century America: A short-lived (and forgotten) marriage. E. KUPFERBERG. Harvard Medical School. Phytopathology 94:S139. Publication no. P-2004-0132-SSA.

Traditionally, historians locate the early development of bacteriology within the disciplines of medicine, public health, and veterinary science. These histories often overlook bacteriology's connections to other fields, including soil science, dairying, and industrial fermentations. One such forgotten area of bacteriological research in nineteenth century America was the field of plant pathology. Beginning with the work of Thomas J. Burrill on fire blight in the late 1870's, botanists regularly enlisted bacteriological methods in their investigations of plant diseases. Bacteriology drew further support from champions of the "New Botany", who stressed the importance of laboratory-based experimental studies. By the mid-1890's, agricultural colleges and experimentation stations encouraged their plant pathologists to examine bacterial pathogens. Armed with these technical skills, many of these same researchers ventured into fields beyond plant pathology, publishing bulletins and papers in sanitary science, veterinary medicine, and even public health.

Moreover, they trained an entire generation of bacteriologists who would later pursue work in hospitals, public health departments, and water filtration plants. This close alliance between bacteriology and plant pathology was, however, short-lived. By the 1910's most plant pathologists had resumed their focus on fungal diseases, cultivating a comfortable disciplinary home within mycology. Bacteriology, in turn, emerged as its own discipline in America, relegating the study of microbial plant pathogens to a small portion of the newly established science.

William Farlow Laboratory and the beginning of the U.S. institutional plant pathology cryptogamy. G. DENIS. CNRS, Université de Lille 1 et Université de Lille 3. Phytopathology 94:S140. Publication no. P-2004-0133-SSA.

When Farlow arrived in 1872 in the Strasbourg de Bary laboratory, to be trained in the new methods of the european cryptogamic science, he wrote to Asa Gray: "Here every one thinks that a knowledge of the larger fungi or algae is contemptible. That the only thing worth living for is to study the development of some of the lower forms. A systematic botanist of Fungi or Algae is regarded as a shallow person, of course, without ability." At the end of his trip, he recognized that what is the "most important [he] have learnt the German way of work and managing a laboratory": " you will be disappointed when you again see me in the amount of systematic fungology which I have learnt but […] I have studied development as applied to some of the minute forms." When he was accepted as Assistant of Cryptogamic Botany at the Bussey Institution, he came not only with cryptogamic botany books but also microscopes, microscopic slides and preparations. His laboratoty became an other place where microbiology settled and diffused in the U.S. The story of the settlement of this first institutional cryptogamy in the U.S. we present, is the story of a compromise between what Farlow hoped to do at first (systematic cryptogamy particularly the Algae) and what he learned from the new cryptogamy specially in connection with the new plant pathology, between the upper and minute forms, between the Botanic Department he finally joined in Harvard and the Bussey Institution created to be a High Agricultural School and between the fundamental microbiological studies and the agricultural social demand.

The living soil: Soil bacteria, ecology, and social cooperation. M. FINLAY. Armstrong Atlantic State University. Phytopathology 94:S140. Publication no. P-2004-0134-SSA.

In the late nineteenth century, as German scientists learned that soil bacteria explained legumes' ability to fix atmospheric nitrogen, a new attitude concerning the interplay of business, government, science, and the environment was emerging on both sides of the Atlantic. The discovery sparked several attempts to commercialize these useful microorganisms, as firms offered bottled bacteria that promised to increase yields without chemical fertilizers. In the both countries, entrepreneurial scientists and private firms led aggressive marketing schemes that sought support from such diverse clientele as American experiment station scientists, Prussian bureaucrats, and African-American educators. The controversies that ensued are illustrative of several issues in the history of twentieth-century science and environmental history. Despite difficulties in the laboratory science of soil bacteriology, these firms found a successful market when they framed their ideas in terms of extrascientific value. Notions of beneficial bacteria found a ready audience; the message that the natural world revealed an interdependent system in which mutually beneficial organisms worked together for the good and balance of the whole was comforting news in the rapidly changing social order. Promoters further claimed these bacteria enabled farmers to challenge international fertilizer trusts, to achieve a harmonious relationship with their environment, and to solidify an agrarian vision of independent capitalists free from urban problems.

The great dying on the vine: Scientific responses to the phylloxera grapevine disaster, France 1867–1900. G. GALE. University of Missouri, Kansas City, MO. Phytopathology 94:S140. Publication no. P-2004-0135-SSA.

In the summer of 1867, a few grapevines died in an unimportant vineyard in the southern Rhône Valley. Within two years, with vines dying at an ever-accelerating rate, regional growers realized that a terrible disaster loomed before them: complete destruction of France's southern vineyards by a mysterious new vine disease, a killer of terrifying swiftness and power. What was this unknown disease? how did it kill? where had it come from? and, most importantly, how could it be stopped? Answers to these questions were neither easy nor quick in coming. In the end it took thirty years, and an army of disparate allies—growers, landowners, small holders, politicians, academic scientists in both France and America, private researchers and, finally, amateur practitioners—to bring the ravaging disease to a standstill. And even then the disease was not conquered: we had merely learned to live with it. Most important among the various phases of warfare with the disease was the initial uncovering of its nature and origin. What is surprising today, and typically forgotten, is that it took the combatants, both professionals and amateurs, seven long years of vigorous, sometimes rancorous debate to settle the issue of the nature and etiology of the phylloxera, as the disease came to be called. My presentation examines this debate, attempting to explain how and why it happened. At bottom, I find the debate based in a fundamental disagreement about two competing theories of plant disease, theories which, themselves, originate in two long-opposed models of human disease.

Reconsidering controversy: The Fischer-Smith debate and the development of biological disciplines. C. MATTA. University of Wisconsin-Madison. Phytopathology 94:S140. Publication no. P-2004-0136-SSA.

My paper reevaluates the debate between Erwin Frink Smith and Alfred Fischer over bacteria as a cause of plant disease. Contemporaries and later authors have portrayed the debate as the moment in which an up-and-coming American plant pathology proved victorious over stodgy German plant science. Following C. Lee Campbell's interpretation, I argue that the debate was not an argument about pathogenicity, but rather a disagreement about the quality of scientific proof phytobacteriology (and its research methods) offered. I develop Campbell's analysis by illustrating that the debate was the product of intellectual tension between two related nascent sciences—bacteriology and plant pathology—competing to establish scientific authority. From about 1880 to 1910, biology diverged into a number of research specialties, each with their own scientific curricula and methods. Because laboratory methods and research questions often overlapped, scientists allied with a certain field often found themselves at odds with their counterparts in other fields whose methods and conclusions differed from their own. The Fischer-Smith debate, therefore, is important because it illustrates the relationship between plant pathology, botany, and bacteriology as these disciplines struggled to establish credibility during a time of disciplinary instability. Knowing the historical relationship between these areas of research reminds us that plant pathology is one component of a large network of agricultural and life sciences that shares common practices, knowledge, and goals.

Reality CV: Recent Successful Job Applicants and Field Leaders Tell it Like it is About the Present and Future of Plant Pathology Careers

Science into practice. K. STEOMER. Agricultural Center Limburgerhof. Phytopathology 94:S140. Publication no. P-2004-0137-SSA.

In my career so far I had two very different but equally fascinating and satisfying functions in fungicide research: screening, evaluating and selecting new fungicidal candidates and supporting products with fungicide resistance monitoring. In my opinion to be successful an R&D organization needs a big diversity of different personalities and skills, out of which the following I always felt are very important: people who are able to work perseveringly and very focused (R&D lives from stability and focus) people who initiate changes, search constantly for innovations in the R&D processes (R&D lives from constant change and innovative approaches) people who interact with the markets and strive for the connection between R&D, business and marketing units (connect R&D and the markets) Working in the plant protection industry gives clearly one of the broadest spectrum of functions a plant pathologist can find in one working place/company. A technical solid background enables us to develop careers in R&D (including regulatory functions) or make transitions into marketing and sales. Very scientifically oriented research, lobbying with regulators, developing long term product strategies, managing products in a given country are just few examples of functions plant pathologists carry out in the industry.

From an applied research position in a chemical company into private practice. C. M. BECKER. BAAR Scientific LLC. Phytopathology 94:S140. Publication no. P-2004-0138-SSA.

Applied agricultural research with fungicides, insecticides and herbicides required broad training in scientific disciplines, computers, and understanding the many aspects of agriculture. Efficient collection of data was essential, followed by frequent presentations of summaries to diverse audiences. Current consolidations in the agrichemical industry has created opportunities for private practitioners. All the above requirements are still essential, with

the added requirement of locating customers or clients. Marketing your expertise becomes you most important responsibility. Pros and cons of industry and private practice will be presented.

Plant pathology in a multinational fruit company and for a mid-sized agricultural chemical company. M. D. GROVE. ISK Biosciences Corp. Phytopathology 94:S141. Publication no. P-2004-0139-SSA.

A look at the benefits and disadvantages of working for a food producing company, with an overseas assignment. What is it like to work for a small and medium size agricultural chemical company with a comparison of international versus domestic U.S. assignments. Challenges, advantages and disadvantages to be discussed and compared.

A non-traditional role: Sales and distributor account management. R. SOUFI. The Scotts Company. Phytopathology 94:S141. Publication no. P-2004-0140-SSA.

Sales and distributor account management are career tracks not often considered by graduating students in plant pathology. This presentation gives an overview of the agricultural chemicals market and the relationships between active ingredient manufacturers, distributors and end users. The presentation also outlines the skills and competencies required in a successful sales representative/account manager.

From basic research in a chemical company to being a professor. K. W. SEEBOLD. University of Georgia, Tifton, GA. Phytopathology 94:S141. Publication no. P-2004-0141-SSA.

Many graduates of plant pathology leave the university to take positions in the private sector. The agricultural chemical industry, for example, employs a significant number of plant pathologists in research and development. It is common for new graduates to move into industry, and those on faculty at the university level also have taken jobs with chemical companies. Switching careers from industry back to academia is less common. The motivation, joy, and anguish of making such a career change will be discussed, and differences between these career paths will be highlighted.

Career opportunities in plant pathology extension. L. P. TREDWAY. Department of Plant Pathology, North Carolina State University, Raleigh, NC. Phytopathology 94:S141. Publication no. P-2004-0142-SSA.

Extension is an important component of plant pathology because of its role in delivering research-based information to growers and other end-users. This is also one of the most exciting areas of the discipline. The extension specialist must always be on the "cutting edge" to keep pace with current problems and changes in crop production systems. A unique set of skills are essential for success in plant pathology extension. Extension specialists must be able to communicate and collaborate with a diverse group of people, including growers, county extension agents, other extension specialists, researchers, crop protection companies, and sales representatives. Extension specialists are often responsible for diagnosis of diseases in their respective crops, as well as applied research to develop effective solutions to disease problems. Therefore, excellent diagnostic skills, problem-solving abilities, and a detailed understanding of crop production practices are critical. Many of these valuable skills are not acquired through traditional graduate education programs. Strategies for students interested in careers in plant pathology extension will be discussed.

Life, liberty and the pursuit of gainful employment: A perspective on career opportunities for the budding plant pathologist. R. M. BOSTOCK. Department of Plant Pathology, University of California, Davis, CA 95616. Phytopathology 94:S141. Publication no. P-2004-0143-SSA.

The uncertainties of the job market coupled with the competition for positions, particularly those in the more traditional paths in research and teaching, always have provoked anxiety for new graduates in plant pathology and related disciplines. I do not anticipate the situation will change that much in the near future; these are realities over which we have little or no control. However, there are strategies and techniques that can be helpful in the interview process and in landing a position. I will discuss some of the skill sets and qualities that we look for when evaluating prospective colleagues. I will also offer a personal perspective on having realistic expectations and some thoughts concerning future opportunities in our field.

How I got my job and why I love it. J. D. DOMINIAK. Plant Protection and Weed Management Section, Maryland Department of Agriculture, Annapolis, MD. Phytopathology 94:S141. Publication no. P-2004-0144-SSA.

As a recent master's graduate, I will describe the actions I took to locate positions in plant pathology. Some of these tactics included conducting online job searches, reviewing trade magazines and journals, and consulting with current plant pathologists about potential openings and contacts. As a result of these actions, I went on several job interviews and I will share how I selected the right position for me. In my position at the Maryland Department of Agriculture, I will explain my duties and relate why I love it, despite all the paperwork.

Skills for a USDA career in research and regulatory plant pathology. D. G. LUSTER. USDA-ARS, FDWSRU, Ft. Detrick, MD 21702. Phytopathology 94:S141. Publication no. P-2004-0145-SSA.

New trends in biological research, emphasizing large-scale approaches spanning genomics, phylogeny, evolutionary biology and novel gene/protein discovery will demand new skills from those currently being trained in plant pathology. The need for rapid applications spawned from the massive information generated by these approaches has opened up new disciplines in information management and data mining. The enhancement of biosecurity at federal facilities, new laws regulating microbial pathogens, and expanding requirements for strict phytosanitary controls provide new opportunities for careers in "regulatory plant pathology", requiring a novel combination of basic and applied skills in plant pathology, biosafety and risk management.

Job-hunting after graduation: Beating the odds as an international student. W. G. D. FERNANDO. Department of Plant Science, University of Manitoba, Winnipeg, Manitoba, Canada R3T 2N2. Phytopathology 94:S141. Publication no. P-2004-0146-SSA.

North American universities have experienced a steady increase in the numbers of international students in plant pathology-related graduate programs. For most of these students, getting into the workforce may become quite a challenge once they have graduated, for a number of reasons. This presentation will address some of these issues, and then show how to succeed in the search for high-quality positions in academia, government and industry. The talk will demonstrate how to write the best "Reality CV" and cover letter for a position, reveal some of the myths associated with job-hunting, and include very personal experiences of the author.