

USDA
AGRICULTURAL RESEARCH SERVICE

**NATIONAL PROGRAM 303 –
PLANT DISEASES**

ANNUAL REPORT FY 2010



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Introduction

National Program 303, Plant Diseases, focuses on developing effective disease management strategies that are environmentally friendly, safe for consumers, and compatible with sustainable and profitable crop production. This Agricultural Research Service (ARS) National Program is conducted in cooperation with related research in other public and private institutions. In addition, NP 303 projects are coordinated with those in National Program 301 (Plant Genetic Resource, Genomics, and Genetic Improvement) toward the overall goal of crop improvement through increased resistance to biotic and abiotic factors and increased understanding of host-pathogen interactions.

The overall goal of NP 303 is to develop and improve ways to reduce crop losses caused by plant diseases. These diseases reduce yields, lower product quality or shelf-life, decrease aesthetic or nutritional value, and, sometimes, contaminate food and feed with toxic compounds. Management of plant diseases is essential for providing an adequate and consistent supply of food, feed, fiber, and aesthetic plants. Reducing crop losses has long been a high priority for agriculture and for ARS. Besides the obvious monetary benefits to producers and processors, successful plant health protection is important for maintaining and increasing food supplies with minimal increases in land under cultivation. Additionally, the knowledge and management of plant diseases of quarantine significance are vital, not only for protecting our domestic crops from foreign disease, but also for maintaining and expanding export markets for plants and plant products.

NP 303 is comprised of the following four components:

- *Disease Diagnosis: Detection, Identification and Characterization of Plant Pathogens;*
- *Biology, Ecology, Epidemiology, and Spread of Plant Pathogens and their Relationships with Hosts and Vectors;*
- *Plant Disease Resistance;* and
- *Biological and Cultural Strategies for Sustainable Disease Management.*

Together, these components yield breakthroughs to understand and control plant diseases and to develop strategies for disease management and control that enhance agricultural production and value. During fiscal year 2010, this program produced many important discoveries and advances. Some of these are described on the following pages, grouped by program component, but not necessarily in order of significance:

Component 1. Disease Diagnosis: Detection, Identification and Characterization of Plant Pathogens

A sensitive molecular diagnostics test was developed to detect potato cyst nematodes. Current traditional methods for validation of the presence of potato cyst nematodes in suspected soils have relied upon physical appearance, tests with susceptible potato plants, and DNA sequencing, all of which are time-consuming and expensive to perform. ARS researchers in Ithaca, New York, have developed highly specific and sensitive molecular methods for the differentiation of the two species of potato cyst nematode which currently exist in the United States. These new molecular diagnostic methods offer rapid, specific, and sensitive identification of potato cyst nematode species compared to traditional methods. The high degree of specificity and sensitivity of these methods should permit nematode detection in soil extracts that contain extremely low amounts of nematode DNA. The methods will provide a valuable detection tool useful in nematode regulatory and quarantine programs throughout the United States. A conventional patent application has been filed for the techniques, and the method has been shared with USDA Animal and Plant Health Inspection Service for internal validation.

New sources of resistance to diverse soybean diseases. In collaboration with scientists at U.S. and Canadian universities, ARS researchers in Urbana, Illinois, identified resistance to Asian soybean rust, caused by the fungus *Phakopsora pachyrhizi*, in soybean breeding lines and in commercially available varieties. Because the pathogen mutates to overcome plant resistance genes, identification and utilization of diverse resistance genes for breeding programs is important for long-term management of soybean rust. This research tells growers which commercial varieties can be used against current forms of the soybean rust pathogen and identified new sources of resistance to incorporate into future varieties. This research further identified the regions of the soybean chromosomes containing genes that confer resistance to seed transmission of soybean mosaic virus, as well as commercial soybean varieties with resistance to Phytophthora root rot and stem rot. This information will be used to develop soybeans with resistance to multiple pathogens.

An efficient diagnostic system for sensitive detection of 14 different viruses infecting important crops. The serological method – Enzyme Linked Immunosorbent Assay or ELISA – is the most commonly used technique in plant virus detection. Real-time polymerase chain reaction (real-time PCR) is quickly gaining greater utilization in plant virus detection due to its sensitivity. However, two major factors limit the practical application of real-time PCR in routine virus detection – the slow sample processing and primer specificity. ARS scientists in Charleston, South Carolina, developed a combination of the two methods – immunocapture real-time PCR technology – that allows efficient processing of large numbers of samples for simultaneous virus detection. Accordingly, the industry partner has developed various testing kits based on this technology and started offering a new line of products in 2010. The success of this technology will help U.S. growers acquire timely and accurate information about the virus infection status in

their crop plants. Thus, the appropriate disease management measures may be deployed either to prevent the onset of the diseases or to effectively manage such diseases.

Multiple approaches developed to combat the citrus greening disease. Because of the difficulty in cultivating the bacteria (*Candidatus Liberibacter asiaticus*), presumed to cause Huanglongbing (HLB) or citrus greening disease, and in propagating and maintaining a large number of HLB-infected plants, it is extremely difficult to screen potential chemicals for HLB therapy and control. Recent advances by ARS scientists in Fort Pierce, Florida, in this area include:

- Identifying a combination of two chemicals that eliminates the pathogen associated with citrus HLB in the plant;
- Developing a method for rapid detection of the pathogen; and
- Fully sequencing the genome of the presumed pathogen.

These new developments will greatly facilitate HLB research and the development of new strategies for control of this devastating disease.

A grower-friendly method for detection of Citrus tristeza virus. Citrus tristeza virus (CTV) continues to be a major limiting disease of citrus, and in California, is managed by state and federal regulatory agencies. The mandatory state eradication program has been replaced by a program of selective removal of trees that are only infected with the most severe strains, with the mild strain being far less destructive. However, the state requires all citrus nurseries to propagate virus-free trees, regardless of the strain. ARS scientists in Parlier, California, in partnership with a private diagnostics company, developed a field-deployable, direct tissue blot immunosorbent assay (DTBIA) to detect CTV. This is a simple, sensitive, and cost-effective detection tool which nurseries can use to test their own trees, and has been used to successfully monitor CTV infection in thousands of budwood trees and nursery increase blocks. The availability of this method provides a user-friendly means for growers to meet the rigorous standard of maintaining and selling virus-free stock and propagations. Since CTV is readily aphid-transmitted, all citrus nurseries in California are vulnerable and can now use the DTBIA kit for this same purpose. Before, nurseries had to request regulatory agencies or the University of California to test their trees, which was expensive and not timely.

Advancing bacterial pathogen recognition through genomics. Because of their small size and inability to grow outside living tissue, phytoplasmas (specialized plant pathogenic bacteria) are difficult to identify. ARS scientists in Beltsville, Maryland, have harnessed the power of genomics for plant bacterial pathogen detection. The USDA's Phytoplasma Classification Database, used worldwide by quarantine agencies, diagnostic companies, scientists, students, and faculty, was expanded significantly and made easier to use to aid in the rapid identification and classification of bacterial pathogens, including new species. A new Web-based tool, iPhyClassifier, for the online gene-based identification and classification of bacterial pathogens incorporates carefully curated databases of pathogen gene sequences for up-to-date classification and comparative studies. As a result, regulatory and quarantine agencies can detect and identify

phytoplasmas more readily. This information is also being used to reduce crop losses through production of certified disease-free germplasm.

Transcriptional analysis of genes of a densovirus to identify transcriptional promoters.

Promoters are genomic DNA sequences necessary for a gene to be expressed, and are important research tools for determining which genes are involved in functions such as resistance to diseases. Viruses infecting cells of higher organisms are being explored as sources of stronger, constitutive, organ, or tissue non-specific promoters. While promoter sequences from simian virus 40, cytomegalovirus, and cauliflower mosaic virus, are commonly used in gene expression studies, their availability and intellectual property issues often restrict their use. ARS scientists in Beltsville, Maryland, in collaboration with scientists at Advanced Bionutrition Corp., identified and characterized three promoter elements of *Penaeus stylirostris* densovirus (PstDNV) and found that all of them function in bacteria, insect, fish, and shrimp cells. The researchers also precisely located the start and end sites of the three promoters, and found that the 'left' promoter had the highest activity followed by the 'middle' and 'right' promoters. These promoters can be used in gene expression experiments in *Rhizoctonia solani* or other plant pathogens to elucidate roles of specific genes in plant disease development.

PCR detection of powdery mildew in vineyards. Growers use fungicide applications to control grape powdery mildew pathogens. To improve early detection, ARS researchers in Corvallis, Oregon, developed a quantitative PCR assay and inexpensive spore traps shown to be reliable under commercial conditions for the detection of airborne spores of grape powdery mildew. The team demonstrated the commercial feasibility of using PCR detection of airborne spores of this pathogen to initiate fungicide applications to control the disease. Three years of validation in commercial vineyards saved 2.3 applications per year, or more than \$113 an acre in application costs, without increasing disease development. This demonstrates that withholding fungicide applications until the pathogen is detected in the air is viable commercially. Commercial application of these procedures has the potential to significantly reduce fungicide use and associated applications costs for managing powdery mildew.

*Genetic diversity of *Agrobacterium tumefaciens*.* *Agrobacterium tumefaciens* is a soil-borne bacterium that causes crown gall disease on walnut. ARS scientists in Davis, California, continued efforts to define the genetic diversity of *A. tumefaciens* as a function of geography, host origin, and most importantly, nursery origin. Using PCR, these scientists have enhanced their forensic ability to identify sources of inoculum which aids the industry as it moves towards the cultivation of *A. tumefaciens*-free planting stock and reduction of post plant infections leading to crown gall formation. Identifying the source of infection facilitates development of cost effective control measures.

Genetics of the sudden oak death pathogen. The full genome of the sudden oak death pathogen, *Phytophthora ramorum*, was recently completed and made publically available. Since most genes in *P. ramorum* have not been previously identified, ARS scientists in Davis, California,

analyzed gene sequences that were turned on at four different life stages to better identify their functions. This provided experimental data to estimate the function of about 75 percent of the genes, but also revealed about 2,500 genes which were not identified by gene prediction programs. Similar RNA sequences could not be found in other *Phytophthora* species. Identifying and characterizing these sequences, in addition to the correction of predicted gene models are key to understanding why *P. ramorum* is very different from other species of *Phytophthora*.

Cause of “crumbly fruit” of raspberry identified. The cause of the economically limiting disease called crumbly fruit of raspberry was found to be a previously unknown member of the reovirus group. ARS researchers in Corvallis, Oregon, together with Oregon State University, characterized the virus, developed a detection method, and demonstrated that the virus is transmitted by aphids. The virus test is being used in ongoing clean plant certification for berry crops and virus elimination programs to ensure virus-free plants are being made available to the industry. Because aphids spread the crumbly fruit virus quickly in the field, using virus-free planting stock and aphid control will be required to minimize the impact of this destructive virus on raspberry production in the future.

Component 2. Biology, Ecology, Epidemiology, and Spread of Plant Pathogens and Their Relationships with Hosts and Vectors.

Mobile pathogenicity chromosomes identified that explains how harmless fungi can change into plant killers in wheat and barley scab. Keeping plants healthy is all about keeping one step ahead of the microbes that can harm them. Fungi that cause plant disease have an uncanny knack for changing and overcoming disease resistance genes that scientists work so hard to breed into plants. *Fusarium* species are among the most important pathogenic fungi and include *Fusarium graminearum* that causes wheat Fusarium head blight or scab. Using comparative genomics, ARS researchers in St. Paul, Minnesota, have determined that *Fusarium* fungal pathogens have the unusual ability to pass particular chromosomes between strains and convert harmless fungi into pathogens. This process explains why new strains of fungi can arise quickly and threaten crops. The discovery means scientists can now start working on a means to thwart the trading of chromosomes and the disease-causing ability that comes with them.

Plant virus used to make animal vaccine. Weakened or killed viruses used to make animal vaccines are currently made using animal cells, such as making flu vaccine using chicken eggs. ARS scientists in Beltsville, Maryland, developed a way to make an animal vaccine in a plant using the plant virus Cucumber mosaic virus (CMV). Part of the protein coat of Avian influenza virus (HPAIV) was inserted into CMV. Another plant virus, potato virus X, was used with the modified CMV to cause the plant to make a protein. When this protein was purified from the plant and injected into chickens, the chicken developed an immune response to HPAIV, indicating that it acted like a vaccine. This research also developed a method to use plant viruses

to assemble sub-microscopic, spherical protein “cages” that can be used to develop extremely sensitive diagnostic tools to detect plant and animal pathogens. These techniques can be used to improve production of vaccines and other proteins.

Markers to fingerprint exotic blackberry rust pathogen. In 2005, *Phragmidium violaceum*, which causes blackberry rust, was discovered in the United States for the first time and caused significant economic loss for Washington and Oregon blackberry growers. Understanding the population structure of the pathogen would yield insight into the origin and potential sources of the introduction. ARS scientists in Corvallis, Oregon, developed microsatellite markers for assessing the pathogen’s genetic diversity. The U.S. pathogen population was discovered to be distinctly different from both the European and Australian populations, indicating either that a population of isolates was introduced in 2004 and 2005, or that multiple introductions occurred. These data, coupled with data on the distribution of pathogen, indicate that *P. violaceum* is now endemic to the United States and management practices must be developed to manage the pathogen.

Developing gladiolus plants resistant to cucumber mosaic virus. Cucumber mosaic virus (CMV) is an important plant virus, with over 1,000 susceptible hosts, including gladiolus for which there is no natural resistance. Symptoms drastically reduce yield and include dramatic flower streaking that makes them unmarketable. ARS scientists in Beltsville, Maryland, genetically transformed and selected gladiolus plants that are resistant to infection by CMV. A highly sensitive method for diagnosis of CMV was also developed by these researchers to detect very low levels of infection needed for this kind of plant transformation. This research will facilitate the evaluation of virus resistance in transgenic gladiolus plants to yield improved floral quality and productivity, and methods developed can be transferred to the many crops affected by CMV.

Understanding the changing nature of Cercospora leafspot in sugarbeet. Fungi can develop resistance to fungicides for agricultural use through different means – either through mutations or sexual reproduction, which increases genetic diversity. Leaf spotting diseases caused by *Cercospora* species cause significant losses on numerous crops worldwide. *Cercospora beticola* is responsible for sugarbeet leaf spot, a disease costing sugarbeet producers millions of dollars a year. Previously, *C. beticola* was only known to reproduce asexually. However, field isolates of *C. beticola* are well-known for high levels of variability, particularly with regard to developing fungicide resistance, making disease management more difficult for growers. ARS scientists in Fargo, North Dakota, have characterized the mating type genes of *C. beticola*, genes that are known to be required for sexual reproduction. The research results suggest that *C. beticola* mating type genes are still active and may play a role in sexual reproduction of this fungus. This information is useful for plant breeders when selecting parents for developing disease resistant sugarbeet and is useful for plant pathologists to help explain how *C. beticola* is able to gain resistance to fungicides.

Migration of Phytophthora ramorum. *Phytophthora ramorum*, the cause of sudden oak death, has been reported in ornamental nurseries on the west coast of North America from British Columbia to California. Long distance migration of *P. ramorum* has occurred via the nursery trade. ARS researchers in Corvallis, Oregon, studied migration and genetic diversity of *P. ramorum*. This analysis provided evidence of four global migration events, of which three occurred into North America. This work highlights the repeated global migration of this pathogen and identified pathways of migration into the United States that can be managed.

Root to root spread of the sudden oak death pathogen Phytophthora ramorum was documented under flooded conditions. Sudden oak death is established in California and Oregon, but not yet established in the eastern United States. When sprouted acorns were exposed to different concentrations of spores of *P. ramorum*, as little as one spore per milliliter caused infection within 24 hours in all four eastern U.S. oak species tested. Forty-six plant species of economic or ecological importance to the eastern United States were evaluated for their ability to support root infection and production of *P. ramorum* spores. Many of these species supported higher populations of the pathogen than a highly susceptible control species, *Viburnum tinus*. This information will be valuable for risk evaluation and regulatory decision-making.

Source of blueberry scorch virus dissemination. *Blueberry scorch virus* (BIScV) was appearing in newly planted blueberry fields in a pattern atypical for normal spread by the insect vector of this disease. ARS scientists in Beltsville, Maryland, determined that blueberries propagated at nurseries can be infected with BIScV, but not show any symptoms. The infected plants are then shipped to farms where the virus symptoms appear within the next 2-5 years. This prompted a state-wide nursery inspection system to identify infected source plants and prevent further distribution.

Component 3: Plant Disease Resistance.

Century-old mystery of the life cycle of wheat and barley stripe rust solved and protected wheat varieties released. Stripe rust (also called yellow rust) of wheat and barley causes significant wheat and barley crop losses worldwide, but the life cycle of the rust fungus has long baffled scientists. ARS scientists in St. Paul, Minnesota, made the first identification of an alternate host for any type of stripe rust pathogen by demonstrating that several species of barberry serve as alternate hosts for the cereal rust. Stripe rust is known to be one of the most variable cereal rust pathogens and this discovery suggests that recombination on susceptible barberry species is playing a key role in contributing to pathogen variability. This information can assist crop breeders in developing entirely new strategies to protect cereals from stripe rust losses. Another advance in cereal stripe rust protection was made by ARS researchers at Pullman, Washington, who exploited stripe rust resistance assays and molecular markers to develop more than 10 new stripe rust-resistant wheat varieties in partnership with ARS and land grant university wheat breeding programs.

New varieties of table grapes and raisins with high fruit quality and Pierce's disease resistance. Introduction of Pierce's disease resistance from wild grape species into table grapes and raisins results in small berries and poor fruit quality. ARS scientists in Parlier, California, have used traditional breeding techniques to generate desired *Vitis vinifera* table grape and raisin varieties that retain Pierce's disease resistance from the less desired grape, *Vitis arizonica*, but that have fruit of high vinifera quality. The current advanced selections will be developed into new table grape and raisin cultivars with Pierce's disease resistance.

Genetic diversity in reniform nematode populations described using microsatellites. Understanding variability in the pathogen population is important not only to improve our understanding of the basic biology of this organism, but also to assist in tracking how the pathogen responds to various management practices and determine the origin and spread of the nematode in the United States. Using six populations of reniform nematode from four states, ARS researchers in Stoneville, Mississippi, developed 156 molecular markers that will enable detection of the genetic variability within this species. Twenty-two of these markers were sensitive enough to detect differences among three populations collected from the same location in Mississippi. This information is being used by plant breeders to develop nematode-resistant plants.

Diagnosis and sequencing of Liberibacter species associated with citrus and potato diseases. Huanglongbing (citrus greening) is the most damaging disease threatening citrus production worldwide. ARS scientists in Parlier, California, have developed a DNA-fingerprinting technique able to distinguish strains of the presumptive pathogen, *Candidatus Liberibacter asiaticus*. Application of the sequence information toward diagnostics will facilitate monitoring potential spread of the pathogen to citrus production areas where huanglongbing disease is not currently known to occur. A related organism associated with zebra chip disease of potato (which renders tubers unsuitable for processing) was sequenced by ARS scientists in Parlier. These sequences will facilitate diagnostic assays and potentially lead to identification of new targets for development of disease resistance.

New approaches to controlling nematodes in watermelon through grafting. Root-knot nematodes (*Meloidogyne* spp.) cause extensive damage to root systems of watermelon and many other vegetable crops, resulting in significant yield losses. Aside from soil fumigation, which continues to have limited use, host plant resistance is the preferred means of control, if available. In recent years, the U.S. watermelon industry has developed an interest in planting grafted watermelons for imparting resistance to soil inhabiting pests and diseases. ARS scientists in Charleston, South Carolina, have identified wild watermelon (*Citrullus lanatus* var. *citroides*) lines resistant to root-knot nematodes. These lines were evaluated as rootstocks for grafted watermelon in field tests in South Carolina and Florida. The root-knot nematode-resistant lines performed well as rootstocks, and produced high yields for the grafted watermelon plants. These wild watermelon rootstocks could be a useful alternative to soil fumigation with pesticides, such

as methyl bromide, for managing root-knot nematodes in watermelon fields and in organic production. Also, they can be useful for seed companies interested in developing rootstock varieties for grafted watermelon.

Component 4: Biological and Cultural Strategies for Sustainable Disease Management

Use of new molecular probes to screen for virus resistance in walnut. In California, black line disease (caused by a virus) adversely impacts walnut production and longevity of some orchards. ARS scientists in Davis, California, have incorporated a resistance gene from black walnut into English walnuts and developed virus resistant backcrosses for release in areas with high incidence of black line disease. The researchers developed and optimized a molecular assay, based on this gene, to identify virus resistant germplasm. In collaboration with the University of California at Davis, more than 700 saplings from different backcrosses were screened and trees potentially resistant to the virus have been identified for field testing.

Identification of the causal agent associated with the almond brownline disease. Almond brown line disease was discovered in California in the 1990s as a graft union disorder in almonds grown on plum rootstock in orchards on marginal land. When the trees are infected by peach yellow leafroll phytoplasma, they develop a brown necrotic line at the graft union resulting in tree death. It has been difficult to prove the association of phytoplasma in infected almond trees because of non-availability of a suitable detection assay. ARS scientists in Davis, California, developed a molecular assay and successfully detected this phytoplasma in almond extracts. This assay will be used to monitor the trees in a commercial orchard impacted by almond brown line disease.

Avoiding phytotoxicity of mustard seed meals to crop plants. Mustard seed meal contains naturally occurring chemicals that make them of interest as management agents for weeds and soilborne pathogens. Previous studies indicated that seed meal from two species of mustard, *Brassica juncea* and *Sinapis alba*, are toxic to nematodes. Because of the potential for phytotoxicity of these meals, it is advantageous to know the amount of time required between mustard seed meal application and planting of a crop to avoid phytotoxicity. ARS scientists in Beltsville, Maryland, and Corvallis, Oregon, tested the meals alone and in combinations to determine toxicity to pepper seedlings and to germinating lettuce seeds. Treatment with *B. juncea* seed meal tended to be the least toxic overall to pepper seedlings, indicating that nematotoxic rates of *B. juncea* could be applied relatively close to the time of pepper transplant. This research is valuable to scientists optimizing the use of seed meal amendments for managing plant-parasitic nematodes without toxicity to crop plants. Growers eventually will benefit from the new nematode control practices.

Resistance to Eutypa dieback identified in grapevines. Eutypa dieback, caused by the fungus *Eutypa lata*, impacts all vineyard production systems (wine, table, raisin, and juice) and affects all U.S. grape-growing regions, with net income losses of \$260 million annually for wine grapes

alone. Given that preventative sprays must be used each year throughout a lengthy period of susceptibility, coupled with the fact that infections are chronic once established, disease-resistant grape varieties are highly desirable as an effective, long-term, and environmentally sensitive solution to fungicides. ARS scientists in Davis, California, identified Eutypa-resistant grape hybrids by screening plant material from the ARS National Clonal Germplasm Repositories in Davis, California, and Geneva, New York. These results are based on a greenhouse experiment carried out by inoculations of a virulent strain of the fungus to non-grafted vines, including seven commercial varieties of *Vitis vinifera* and seven experimental varieties of *Vitis* species or interspecific hybrids, the latter of which are components of breeding programs in California, New York, and Australia. As these Eutypa-resistant hybrids are also resistant to the destructive and widespread fruit and foliar pathogens powdery and downy mildews, establishing vineyards with such varieties, or using them to breed other varieties, is likely to reduce pesticide use and, thus, lessen the impacts of grape production on human health.

Crown gall resistant walnut germplasm. Crown gall is an important disease in walnut, and resistant germplasm is needed. ARS scientists in Davis, California, found under greenhouse conditions that durable crown gall resistance is present in *Juglans microcarpa*, *J. ailantifolia*, *J. mandischurica*, and *Pterocarya* accessions. Over 1,500 individual greenhouse propagated *Juglans* seedlings were screened for crown gall resistance. Approximately 15 percent of the rooted dormant cuttings from mother trees exhibiting crown gall resistance continue to exhibit resistance. New directed crosses were made generating interspecific hybrids which exhibit various degrees of crown gall resistance. Development of crown gall resistant stock would save \$20 million in annual losses to the walnut industry.

Interaction between lesion and root-knot nematodes in peach. A 26-month field study was conducted at ARS Byron, Georgia, to determine the interaction between the root-knot and lesion nematodes as related to peach tree growth. Results indicate that the root-knot nematode suppressed tree growth in peach more than lesion nematode. These data provide useful insights into the need for developing appropriate root-knot nematode management strategies in peach.

Biocontrol agent for fire blight improved. Biological control of fire blight of apple and pear with beneficial microorganisms is a viable alternative to the use of antibiotics, which have become less effective due to development of resistance in the causal organism. One effective biocontrol agent, the bacterium *Pantoea agglomerans* strain E325, was originally discovered by ARS scientists in Wenatchee, Washington, and is now commercially available. In efforts to improve the production and tolerance of this organism to dehydration and related stress factors, salt levels were increased in the growth medium. As a result, the organism survived at significantly higher levels during freeze drying and subsequent storage prior to application, and in some cases, established better on floral surfaces under dry conditions. The research led to commercial modifications and an improved biological control product for fire blight management.

New Phomopsis species identified from grapevine. Phomopsis cane and leaf spot, caused by the fungus *Phomopsis viticola*, is a destructive foliar disease in eastern North American vineyards, and its control requires a minimum of four preventative sprays per growing season. Infections can become so numerous on leaves and stems that the vine drops many of its leaves or the stems are girdled (and the fruit subsequently shrivels). ARS scientists in Davis, California, and Beltsville, Maryland, in cooperation with scientists from University of California-Riverside, and Cornell University, Geneva, New York, identified two new species not previously found on grapevines: *P. fukushii* and *Diaporthe eres*. The researchers are currently evaluating the virulence of these new species, in comparison to *P. viticola*, on potted plants in the greenhouse. If these new species infect grapevines via pruning wounds, then standard controls aimed at preventing infections of leaves and stems may not be effective against pruning wound infection.