# USDA FY 2010 AVOIDING HARM FROM INVASIVE SPECIES (USDA Do No Harm 2010 Report)

## A USDA Report to the Invasive Species Advisory Committee and the National Invasive Species Council by Hilda Diaz-Soltero, USDA Senior Invasive Species Coordinator

March 14, 2011

There are eight U.S. Department of Agriculture (USDA) agencies that work on invasive species issues: the Agricultural Research Service (ARS); Animal Plant Health Inspection Service (APHIS); Cooperative State Research, Education and Extension Service (CSREES) (now the National Institute for Food and Agriculture (NIFA)); Economic Research Service (ERS); Farm Service Agency (FSA); Foreign Agricultural Service (FAS); USDA Forest Service (FS) and Natural Resources Conservation Service (NRCS).

Previous USDA Do No Harm Reports cover: (1) fiscal year (FY) 2004 activities; (2) FY 2005 activities for ARS, APHIS, CSREES, ERS and NRCS (first report dated October 2004); (3) FY 2005 activities for the Forest Service (report dated February 2005); (4) FY 2006 activities for ARS/NAL, CSREES, ERS, NRCS and USFS (report dated March 2007); (5) FY 2006 activities for APHIS (report dated August 20, 2007); FY 2006 activities for ARS (report dated September 22, 2007); and FY 2007 activities for APHIS, ARS,ARS/NAL, APHIS, CSREES, ERS, FAS, FS and NRCS (report dated 20 March 2008); FY 2008 activities for APHIS, ARS, ARS/NAL, APHIS, CSREES, ERS, FAS, FS and NRCS (report dated March 3, 2009); and FY 2009 activities for ARS, ARS/NAL, APHIS, NIFA, ERS, FS and NRCS (report dated February 17, 2010) .

**This is the tenth "USDA Do No Harm Report" to the Invasive Species Advisory Committee and the National Invasive Species Council. It covers the FY 2010 activities for ARS, ARS/NAL, APHIS, NIFA, ERS, USFS and NRCS. The report is dated 14 March 2011.**

The report is divided by agency activities. Each agency will report on:

a) Invasive species program activities the agency is carrying out to do no harm;
b) The way in which, when the agency carries out other programs activities, they are also designed and implemented to do no harm;
c) Activities that are doing harm and future actions the agency will take to change the activities so that they do no harm.

Within the above categories, the agency will include its own agency activities, as well as activities where the agency is coordinating and/or collaborating with another federal agency, per the mandate of the Invasive Species Executive Order (EO 13112).

## I. USDA Research Agencies:

### A. Agricultural Research Service (ARS)

The **Agricultural Research Service** (ARS) Agricultural Research Service (ARS) is the principal in-house research agency of the USDA. With a staff of over 8,500 employees, ARS carries out research at over 100 laboratories throughout the Nation and in several foreign countries. ARS research is organized under four broad categories: Animal Production and Protection; Nutrition, Food Safety, and Quality; Crop Production and Protection; and Natural Resources and Sustainable Agricultural Systems. Pest management, including invasive species, is a major research component across all these areas. Research infrastructure dedicated to pest management includes personnel and facilities in domestic and foreign laboratories that also provide support to other agencies, organizations, and state governments. ARS is committed to performing its research programs and projects in a manner that does not cause or promote the introduction or spread of invasive species in the U.S. or elsewhere, ensuring that all feasible and prudent measures are taken to minimize risk of harm.

### 1. Activities that do no harm

#### A. **Informational Activities.**

- **e-Government and Public Communication Initiatives.** USDA's National Invasive Species Information Center at the National Agricultural Library (NAL) maintains and manages the www.invasivespeciesinfo.gov Web site as a reference gateway to information. The Center and its Web site serve a broad customer base, from students, to farmers, researchers, and government officials. Special attention is given to serve the information needs of the professional media.

- The impact of the NISIC program continues to increases as well as visibility as an authoritative USDA resource for Invasive Species. The www.invasivespeciesinfo.gov Web site is frequently cited as a good source of information in many news articles. The Web site consistently is ranked highly in all major search engines.

- In FY 2010:
    - More than 366 requests for information were answered (+15% increase from previous year).
    - Web Statistics:
        - NISIC Web site – 2.6 million page views (+13% increase from previous year), 852K unique visitors (+8% increase from previous year)
        - NISIC What's New Blog – 173K page views (+23% increase), 114K unique visitors (+1% increase)
        - ITAP Web site – 12K page views (+234% increase), 4K unique visitors (+287% increase)

- Responded to various media requests for information related to invasive species, including:
    - History Channel – producing a documentary on invasive species for animals and plants in the U.S.
    - Long Haul Productions - producing a documentary on the Asian carp for public radio.
    - World Fishing Network / WFN News – looking for information for Asian carps to help inform the public understand the threats of Asian carp.
    - Mountain Lake PBS – documentary on invasive species using Lake George Basin as microcosm for Great Lakes.

- Outdoor life Magazine – writing article on invasive species.
- Discovery Channel - various questions about states having problems, rate and history of introduction, etc.
- ABC 20/20 and Primetime – looking for invasive species story ideas. Discovery Channel (TV producer in England) – producing TV series about invasive species; the damage they are doing, what's being done to tackle them, what the general public can do to help, etc.
- WMCT-TV in Marlborough, Massachusetts seeking experts for a television program concerning the threat of invasive species, specifically the Asian Longhorn Beetle.

- ForeSee Customer satisfaction results show a consistent improvement in customer satisfaction with NISIC rated as one of the highest rated Web sites at NAL. Satisfaction scores for NISIC appear to be trending very gently upward. Satisfaction score tend to impact future behaviors, including likelihood to recommend the site to others, and to return to the site.

- Consistent #1 ranking in search engine results for "invasive species" as well as many other pages within our site for specific species information.

- FY 2010 NISIC Information Products and Enhancements:
  - **ARS Promo Box** -- Created a promo box for ARS's Research on Invasive Species on NISIC's home page as per John Lydon's (ARS National Program Leader) request. Edited and reorganized existing ARS information to help users find related ARS program information on our site.
  - **Cross-cutting Issues for ARS** -- Researched information on the linkages between invasive species and global climate change, food security and forest or wildfires. Revised the current Hot Issues section pulling together resources and creating RSS feeds on each of these subjects.
  - **Twitter** -- Continued using Twitter (InvasiveInfo) incorporating our Invasive News feed and What's New feed automatically, as well as adding custom tweets. At

the end of FY2010, InvasiveInfo had 374 followers with 46 lists following our Twitter. In many instances, our content was retweeted on our followers' pages.

- **Social Bookmarking** -- Continued using a social bookmarking widget on our site pages which allows users to easily add pages on our site to various common social bookmarking sites. This utility will enable us to monitor additional statistics and extend our outreach. Approximately 200 users shared our Web site information via emails, bookmarks, and RSS subscriptions.
- **New Content** -- Frequently added new relevant content for many site topics including: invasive species bills, federal and state press releases, management plans, grants and funding opportunities, conferences and events, education for professionals, specific profiles resources, and much more. Developed 10 new species profiles. As a result of comments from ForeSee survey responses, provided more state specific information.
- **RSS Feeds** -- Maintained our customized RSS feeds for What's New on Our site, Invasive Species News, Invasive Species Related Grants (Grants.gov), Invasive Species Journal (Invasive Plant Science and Management),and various Emerging Issues feeds. Hundreds of Subscribers receive our various daily email updates; users include staff from various Federal and State agencies, universities and school systems.

- **National Invasive Species Council Support.**
  Continued to support the activities of National Invasive Species Council by posting relevant information and as requested by Hilda Diaz-Soltero, USDA Senior Invasive Species Coordinator (conferences, federal register notices, Invasive Species Advisory Committee information, etc), as well as additional information from the Federal Agencies representing the National Invasive Species Council.

- **Other e-Government and Public Communication Initiatives.**
  Invasivespeciesinfo.gov Web site links: USDA's National Invasive Species Information Center Web site links to the 13 Federal Agencies that are members of the National Invasive

Species Council, as well as links to the many Agency specific programs and resources relevant to invasive species issues. NISIC also includes extensive resources for State, Professional and Non-Profit, and International programs with an interest in the prevention, control, or eradication of invasive species.

- Information management support to ITAP: The National Agricultural Library (NAL) provides information management support for the Federal Interagency Committee for Invasive Terrestrial Animals and Pathogens (ITAP), a Federal scientific and technical interagency advisory group. This includes:

    - In FY 2008, NAL launched the www.itap.gov Web site for the interagency committee; NAL continued to support ITAP.gov in FY2010.
    - Supports SharePoint a secure Web-based internal communication platform.
    - Listserv for committee-wide communication.

B. **ARS Research Activities.**

**Plant Diseases: Detection, Identification, Characterization, and Monitoring**

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.

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.

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

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.

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.

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.

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.

New germplasm and genetic resources developed to protect wheat and barley from the Ug99 stem rust. Wheat and barley germplasm with resistance to the virulent Ug99 stem rust strain is urgently needed to protect the global grain supply. ARS researchers in Raleigh, North Carolina; Aberdeen, Idaho; and St. Paul, Minnesota, evaluated 4,000 wheat and barley varieties and germplasm lines in Njoro, Kenya, for resistance to Ug99 stem rust. Lines were submitted from more than 25 public and private sector U.S. breeding programs. ARS researchers in Raleigh also developed 750 wheat lines with stem rust genes combined in two, three, four, and five gene stacks (or pyramids). These lines also have gene combinations for leaf and stripe (yellow) rusts. The researchers in Raleigh also developed and distributed 30 advanced lines of wheat incorporating multiple-gene resistance to stem rust race Ug99 to wheat breeders in 32 countries in cooperation with the International Wheat and Maize Improvement Center in Mexico. This information will enable breeders in the United States to identify and deploy resistance to Ug99 stem rust in advance of the pathogen ever arriving in the United States. These lines will greatly aid U.S. and international wheat breeders develop better worldwide resistance to stem rust race Ug99.

Description of the fungus *Dolabra nepheliae* on rambutan and lychee. Fungi are a large and diverse group of organisms that cause serious diseases of crop and forest plants. Accurate knowledge of fungi is critical for controlling the diseases they cause. Rambutan and lychee are tropical plants that produce delicious edible fruits. A little known fungus causes a canker disease, known as corky bark disease, in rambutan and lychee in Hawaii and Puerto Rico. *Dolabra nepheliae* has not been reported on Litchi and Nephelium in Hawaii and Puerto Rico prior to 2007. The fungus, originally described from Malaysia and has been reported from Australia, causing this disease was described and illustrated by ARS researchers in Beltsville, Maryland, Mayaguez, Puerto Rico, and Hilo, Hawaii. The fungus's relationship to other disease-causing fungi was also determined. This new knowledge will allow plant pathologists to accurately identify the cause of this disease of specialty crops. Knowledge of this fungus is useful to plant regulatory officials working to control the spread of this disease.

## Insects: Detection, Identification, Characterization, Monitoring, and Control

Insect and mite systematics help safeguard the Nation's agriculture. Invasive species cause hundreds of billions of dollars in losses in the United States each year. Systematics collections are essential for addressing these threats. During the past year, ARS scientists in Beltsville, Maryland, used these insect and mite collections to conduct 46,000 identifications, including over 6,000 considered urgent by the USDA Animal and Plant Health Inspection Service, from specimens collected at U.S. ports. The researchers produced electronic identification tools for invasive fruit flies; descriptions of new parasitic wasps that attack leaf-mining flies and other wasps used for biocontrol of the invasive weed, Old World climbing fern; and identification of flea beetles used for biocontrol of other invasive weeds. In addition, this research produced knowledge of moths that is assisting in the Discover Life in America effort documenting all life in the Great Smoky Mountains National Park for the purpose of biodiversity education. The scientists also are discovering clues to host-parasite evolution through leaf-mining fly systematics, and conducting extensive biological and ecological studies of a new

parasitic wasp found on the important invasive emerald ash borer. These applications are being used to prevent the introduction of new invasive species and manage established ones.

Brown marmorated stink bug can cause significant damage to apple and peach. The brown marmorated stink bug is a recently introduced invasive pest in eastern North America that has begun to cause economic damage and is a household nuisance. ARS scientists in Kearneysville, West Virginia, showed that early season damage is greater than 20 percent in apple and 50 percent in peach; in both unsprayed and sprayed orchards. The insect growth rate was also confirmed. Damage estimates and sampling will enable stink bug monitoring for determination of when damage levels to fruit are imminent. Application of these data to previously developed growth rate models will predict when the brown marmorated stink bug is at the proper growth stage to manage.

Ecological clues to stink bug control in the South. Three major stink bugs (southern green, brown, and green) affect key southern crops such as cotton, corn, and peanut; which are often grown in rotation. Ecological and behavioral studies by ARS researchers in Tifton, Georgia, have shown that natural enemies of stink bugs, including tiny parasitic flies, predatory fire ants, spiders, and others are responsible for significant control, particularly when border vegetation is conserved, and that early planted corn suffers less stink bug damage than late planted corn. Overall results suggest that there are crop-specific predator species that are able to cause high mortality of stink bug egg masses. This work will be employed by those combating the newly invasive brown marmorated stink bug. In addition, the researchers demonstrated for the first time that the southern green stink bug can be trapped with its pheromone. The results suggest that stink bugs trapped with a pheromone blend from all three stink bugs has the greatest potential for detecting all of the stink bugs in diversified agro-ecosystems.

New plant acid-based varroa mite treatment developed. Varroa mites are an external parasite of honey bees and the major cause of colony losses throughout the United States. ARS scientists in Tucson, Arizona, created a formulation using plant acids that is highly effective in reducing varroa mite populations in bee colonies. These

plant acids are food grade compounds and are on the FDA's "generally recognized as safe" list. The product delivery system causes bees throughout the colony to have levels of the product that result in varroa mite mortality in less than 48 hours and does not cause mortality in either adults or immature life stages nor disrupt queen egg laying or colony growth. The product does not accumulate in the wax comb and, in most cases, was not found in honey samples; when it was found it was in very low amounts, less than 100 parts per billion. The product was developed under a Cooperative Research and Development Agreement (CRADA) and is in commercial production under the name HopGuard. This product should significantly improve mite control in a manner that is non-toxic to the bees.

New control solutions for aphids developed based on neuropeptide hormone technology. Many aphid species entering are vectors of new diseases and cause hundreds of millions of dollars of crop damage yearly. ARS researchers at College Station, Texas, in cooperation with British colleagues, developed an entirely new approach for the control of pest aphids. The technology is based on developing versions of natural aphid hormones (known as neuropeptides) that resist metabolism (inactivation) by natural aphid body enzymes. Natural neuropeptides in aphids and other insects regulate critical life processes such as water balance and digestion. Some of the neuropeptide "mimics" developed by this work match or even exceed the potency of current insecticides used in aphid control. While the development of commercially viable neuropeptide technology for aphid control has not yet been realized, this accomplishment is moving the work forward, and is catalyzing related work by other scientists in industry, academia, and government.

A novel approach for detecting Russian wheat aphid infestations in wheat fields. The ability to quickly categorize pest status in large wheat fields is critical to facilitating timely application of control measures, but there are no efficient methods available to sample large fields at this time. ARS researchers in Stillwater, Oklahoma, with collaborators, developed remote sensing technology to detect and monitor infestations of Russian wheat aphids in production winter wheat fields. Over the duration of this project, airborne multispectral imagery was acquired from numerous production wheat fields in

western Oklahoma and southeastern Colorado, then processed and analyzed using both standard and novel analytical methods. Stress caused by the Russian wheat aphid could be detected with multi-spectral imagery of infested wheat fields. Using a combination of spectral information combined with spatial images of stressed plants, stress caused by Russian wheat aphid could be differentiated from non-stressed fields and fields that were stressed by common environmental factors such as drought. In the future, the ability to categorize the pest status for fields using airborne multi-spectral remote sensing will facilitate timely application of control measures without the need for expensive and time-consuming within-field pest scouting.

Control strategy mitigates the threat of the invasive Argentine cactus moth in the United States and eradicates the pest in Mexico. Since its detection in south Florida in 1989, the Argentine cactus moth has expanded its range each year along the Atlantic Coast and west along the Gulf Coast to the barrier islands of Mississippi. This moth has become an imminent threat to many *Opuntia* cactus species which are valued as food, forage, wildlife habitat, and a major plant group contributing to ecosystem structure and biodiversity. ARS researchers in Tifton, Georgia and Tallahassee, Florida, in collaboration with the USDA Animal and Plant Health Inspection Service (APHIS), developed and refined survey methods and control tactics, using field sanitation combined with sterile insect releases, along the leading edge of the invasion and at new outbreak locations. With the cooperation of ARS, APHIS, and SAGARPA (Mexico's department of agriculture), Mexico continues to adopt and implement these methods and tactics in the operational program that is part of an ongoing United States-Mexico bi-national campaign against this invasive pest. These actions have eradicated or greatly reduced established populations of this pest on the Alabama and Mississippi barrier islands and the islands off the coast of Quintana Roo, Mexico, as well as mitigated the further westward expansion of pest populations along the Gulf coast. This is the first time any moth pest has been eradicated from a country in the Western Hemisphere.

Chemical pesticides for growers of nursery and floral crops. Horticultural plants shipped into the US can serve as a source of invasive species. The ornamental horticulture industry is vast and the

plants are grown under a number of conditions such as nurseries, greenhouses, and tree farms. The plants can be in beds, containers or in-ground. The growers are involved in a number of diverse markets including flowers, bulbs, houseplants, perennials, trees, shrubs, nonbearing fruit trees, and others. Treating such an enormous variety of plants with pesticides presents a challenge for crop safety, so considerable that effort must be spent in developing phytotoxicity data so that pesticide manufacturers will add these crops to their labels. In 2010, ARS scientists established 210 pesticide/crop combinations at field locations in six states (South Carolina, Mississippi, Georgia, Washington, Texas, and Ohio) to treat with pesticides and evaluate them for crop safety. A select number of these combinations were also evaluated to see how well the pesticide performed against the target pest. ARS contributed data toward the registration of uses for 112 crops and 23 pesticides that are now available to growers of florist and nursery crops to reduce losses from pests.

Natural biological control of the imported fire ant. Fire ants, accidentally imported from Argentina in the early 20$^{th}$ century, are unusually abundant in the United States, because they have escaped their natural enemies left behind in South America. Scientists in Gainesville, Florida, obtained approval from the North American Plant Protection Organization and USDA-APHIS regulators to release a new species of phorid decapitating fly (*Pseudacteon cultellatus*) as a fire ant biocontrol agent. This new species of fly specializes on attacking the smallest sizes of fire ant workers, which are most abundant in multiple-queen fire ant colonies. This preference is especially important because multiple-queen fire ant populations average two to three times the densities of regular single-queen fire ant populations and are therefore a substantially greater pest of homes, agriculture, and the environment. Another example of significant progress in development of biological control was the comparison of a new fire-ant virus, SINV-3, from American and Argentinean populations. Working with the USDA ARS South American Biological Control Laboratory, scientists sequenced the SINV-3 genome in entirety from an Argentinean source and compared it with the genome sequence found in U.S. populations. The Argentinean variant had a different genomic architecture and may exhibit different virulence levels compared with the U.S. variant.

ARS scientists also showed that more recent infestations of fire ants in Australia, China, and Taiwan probably originated from the United States rather than from South America. These subsequent invasions of fire ants may have been facilitated by genetic pre-selection in the United States. This knowledge is valuable because the same biological control agents that are effective in the United States are likely to be helpful in Asia and Australia. Establishment of a complex of biological control agents in the United States and other countries where the fire ant has invaded will likely establish the system of natural controls necessary to reduce the impact of this pest.

Discovery of termite enzyme systems for pest control and bioenergy. There are few molecular target sites for the development of new termite control chemicals. Scientists in New Orleans, Louisiana, have cloned two enzymes from the Formosan subterranean termite and expressed them in a bacterial expression system. Incubation of the two enzymes together with cellulose results in digestion of the cellulose to glucose. This will allow us to test a variety of possible inhibitory compounds against the digestive enzymes necessary for the termites as a possible wood preservative or termite control agent. An additional outcome for this research is the possible use of these enzymes for the conversion of biomass into biofuel.

Development of sterile insect technology (SIT) to control the light brown apple moth. The invasive light brown apple moth (LBAM, *Epiphyas postvittana*) is a serious pest that attacks many types of plants and poses a threat to both agricultural and non-agricultural areas across the United States. SIT is an effective in pest control strategy used to control various economically important insect pests and could be one technology to help stem the invasion of this damaging pest. ARS scientists at Hilo, Hawaii determined an effective sterilizing irradiation dose range for LBAM adults for use in an SIT program. This information is essential for the development and implementation of an environmentally safe and effective SIT program for IBAM and will be of use to agriculturalists interested in such an effort.

Mating Affects Female Attractiveness in Lygus. Lygus bugs are polyphagous pests that pose a significant threat to a number of economically important crops including cotton and the emerging

biofuel crops, camelina and canola. After mating, Lygus bug females enter a refractory period in which they become unresponsive to male mating attempts. ARS scientists Maricopa, Arizona showed that male Lygus bugs transfer a compound to females during mating which renders them less attractive to other males. The compound, found in the medial and lateral accessory glands and in the spermatophore (sperm packet) provided to the female, has relatively low volatility. Topical application of the compound renders virgin females unattractive to males, thus disrupting mating. Knowledge of existence of this compound and its impact on mating can be used to develop mating disruption products that, when applied early in the season to limit field populations of Lygus bugs, would reduce the need for use of broad spectrum pesticides.

Fungal pathogen for control of potato psyllid. Potato psyllid is a highly invasive pest of potatoes, tomatoes, and peppers. Although pathogens in psyllids have been described, no fungi have suitable as biological agents for psyllid control have been reported. In an initial screening of fungal pathogens of psyllids for their potential as biological control agents, ARS scientists at Wapato, Washington found four isolates of fungi that caused 91-99% mortality in psyllid adults and nymphs within 4 days after treatment. Of these, two isolates had good biological activity on psyllids under field conditions, where reductions in psyllid numbers were accompanied by increases in tuber yield. The information from this study will be useful to agricultural practitioners interested in controlling psyllids with no or reduced chemical inputs.

Identification of psyllid sex pheromones. Psyllids are major pests of a number of important crops. The main damage caused by psyllids is the result of diseases that they transmit to the plant while feeding. Their detection and control is often accomplished through the use of traps baited with sex pheromones. It has not been possible to take this approach for the control of the potato psyllid, which also feeds on other Solanaceous plants such as tomatoes and peppers, or the pear psyllid because sex attractants for these psyllids were not know until ARS scientists at Wapato, Washington and a University of California chemist teamed up to discovery such compounds. Studies conducted by this research team on volatile compounds produced by females of these psyllid species revealed the presence of chemicals

that attrack male psyllids. While the specific compounds responsible for attracting the male potato psyllid needs to be confirmed, the male-attracting compound produced by the female pear psyllid has be identified. Furthermore, a synthetic version of this compound attracted males pear psyllids in both laboratory and field assays. This is the first identification of a sex pheromone in any species of psyllid. Advances made in these studies could lead to the commercial production of a synthetic attractants for use in monitoring or managing of psyllid populations, and consequently the diseases they spread, in several important crops.

Precision Management of Codling Moth. Codling moth, a major pest of apple and pear, is primarily managed with a series of calendar sprays applied to the entire orchard during the season. To reduce costs and the potential negative impacts to human health and the environment, ARS scientists at Wapato, Washington tested the use of precision agricultural techniques that restrict pesticide applications both spatially and temporally based on male and female insect density thresholds. Insect traps baited with lures attractive to both sexes were distributed in high density across several hundred acres of pear trees and only the portions of the field where male and female moths were detected above a set threshold were sprayed. Using this approach, management costs for codling moth were reduced 40-60%, and that also reflects a significant decrease in pesticide exposure for applicators and the environment. Adoption of this precision agriculture approach for codling moth in both pear and apples could reduce the acreage treated and management costs throughout the western fruit-growing regions.

Control of Psocids in stored grain. Psocids, or booklice, are emerging pests in stored products, including stored grains, however, recent studies indicate that psocids are tolerant to common insecticides used to control other stored-grain insect pests. ARS scientists in Manhattan, Kansas evaluated several grain protectants registered in the United States for control of different psocid species. These studied demonstrated that control of Psocid adults and progeny production on wheat and rice was best obtained with the insecticide Storicide II® (chlorpyriphos-methyl + deltamethrin), while Actellic® (pirimiphos-methyl) was effective in controlling the pest on

corn. This information will be of value to managers of stored grain in the event that Psocid insect infestations occur.
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Enhancing control of wood boring insects using a new gel formulation of beneficial nematodes. The lesser peachtree borer is a major pest of stone fruits (such a peach and plum). The insect attacks aboveground portions of the tree by boring into the trunk and scaffold limbs. Safe and effective methods of controlling this pest are needed. Beneficial insect-killing nematodes are safe environmentally friendly natural insecticides that are used to control a variety of soil-dwelling pests. However effective above ground applications of the beneficial nematodes to control of the lesser peachtree borer is hindered by the nematode's sensitivity to desiccation and UV radiation. In response to this problem, ARS scientists at Byron Georgia in cooperation with scientists at the University of Georgia and University of Florida developed a novel sprayable gel formulation that protects the nematodes from harmful environmental conditions during aboveground applications. Nematode applications made in conjunction with the sprayable gel resulted in 70 to 100% suppression of the target pest. This new formulation promises to enhance the efficacy of beneficial nematodes in controlling lesser peachtree borer as well as other wood-boring insects and other aboveground pests.

Identification of beetle infestations in palm trees. The invasive red palm weevil (*Rhynchophorus ferrugineus*) is a destructive pest of horticultural and ornamental palm species. Generally, the presence of the weevil goes undetected until the plant is nearly dead. The lack of visible presence of the weevil complicates eradication programs, which often result in the destruction of non-infested trees in areas where infested trees are found. ARS scientists in Gainesville, Florida, developed an acoustic detection method based on signal processing methods that distinguish red palm weevil noises from background noise and sounds produced by many of the other non-pest insects commonly present in the palm trees. This acoustic detection method for the red palm weevil will be used in monitoring and eradication programs in Curacao and Aruba to selectively identify and destroy infested trees, greatly reducing the unintentional

destruction of non-infested trees.

Microbial control of the invasive cactus moth. The invasive cactus moth is a serious threat to native cactus in the southwestern USA and the cactus industry in Mexico. ARS scientists at Tallahassee, Florida in collaboration with scientists at Florida A&M University assessed the use of fungal entomopathogens against the egg and early larval stages of the moth, because these life stages are found outside the cactus pads. Young larvae of *Cactoblastis cactorum* were found to be susceptible to both *Metarhizium anisopliae* and *Beauveria bassiana*, with *M. anisopliae* being extremely virulent. This knowledge will be of value to researchers developing additional biological control strategies for the cactus moth.

Elucidating the chemistry of host finding by the Varroa mite. The Varroa mite is the most important pest of the honeybee industry in the world and no highly effective control strategy exists for this pest. In order to develop a non-pesticide approach to control of the Varroa mite, ARS scientists at Gainesville, Florida investigated the semiochemical communication system used by Varroa mites to invade honeybee larval cells. We identified two semiochemicals that effectively cause these mites to be attracted to empty rearing cells. Filling the atmosphere of bee hive frames with these two compounds confused Varroa mites such that they were unable to find larvae on which to feed. This knowledge will be of benefit in developing non-pesticidal methodologies to protect bee hives from invading Varroa mites.

## Weeds: Detection, Identification, Characterization, Monitoring, and Control

Herbicide selection for Orange hawkweed control. Orange hawkweed is a troublesome invasive weed in pastures and open fields in Alaska. ARS scientists in Fairbanks, Alaska determined that two herbicides, aminopyralid and clopyralid, were very effective at controlling orange hawkweed. Aminopyralid is best used where grasses are the desirable vegetation, whereas clopyralid, which does not control as many broadleaf plant species, is best in areas where maintaining species diversity is desired. These results will be useful to land

managers when selecting herbicides to control orange hawkweed growing in diverse habitats.

Temperature modulated transcriptome of leafy spurge crown buds. Dormancy of vegetative buds is a critical developmental process that allows perennial plants to survive extreme seasonal variations in climate and circumvent weed control strategies. Management of the noxious perennial weed leafy spurge, depends on knowledge about the biology of crown bud dormancy and vegetative reproduction. ARS scientists in Fargo, North Dakota with university partners investigated low temperature induced patterns of gene expression in crown buds of leafy spurge affecting dormancy and flowering.. With the use of sophisticated software programs, these scientists identified central regulators of important biological processes associated with temperature induced dormancy status and flowering in crown buds of leafy spurge; these may serve as candidate genes for future manipulation of plant growth and development. This research will help physiologists identify genes and pathways that underlie dormancy and vegetative reproduction in perennial plants which will in turn help to develop next generation weed management strategies and perennial plant production practices under global climate change conditions.

Determination of life cycle of the arundo leafminer. The arundo leafminer *Lasioptera donacis* (Diptera: Cecidomyiidae) is widespread and abundant in the areas of southern France and Spain. ARS scientists in Weslaco, Texas, in collaboration with ARS scientists at the European Biological Control Laboratory (EBCL) in Montpellier, France, collected arundo leafminers and determined that they completed their life cycle in one month. Adult females lived one to six days and laid eggs in the leaf sheaths of arundo plants, especially in holes made by other insects. Feeding by the larvae and infection by surface fungi caused leaves to turn yellow and die. Studies in southern Spain further characterized the critical factors for arundo leafminer reproduction and efficacy. This work has led to the first successful rearing of a grass-feeding, non-galling cecidomyiid leafminer in quarantine. This success will pave the way for host range and efficacy studies in quarantine, furthering the development of the arundo leafminer as a biological control agent for *Arundo donax* (giant reed) which can outcompete and displace native

vegetation in riparian zones, reduce wildlife habitat, increase fire risks, interfere with flood control, and create a security problem along the southern border of the United States and Mexico.

Aerial release methods for the arundo wasps developed. Ground releases of the arundo wasp *Tetramesa romana* to control *Arundo donax* (giant reed) are not practical in the remote areas of the Rio Grande Basin of Texas because of poor roads and the dense thickets these weeds form; thickets that inhibit uniform dispersal of the wasps. *Arundo donax* can outcompete and displace native vegetation in riparian zones, reduce wildlife habitat, increase fire risks, interfere with flood control, and create a security problem along the southern border of the United States and Mexico. ARS scientists in Weslaco, Texas, in collaboration with APHIS equipment specialists at Moore Airbase, Edinburg, Texas, have developed technology to contain, transport, and release arundo wasps from light aircraft. Specialized cardboard boxes filled with chilled arundo wasps were accurately dropped into the narrow corridor of *Arundo donax* thickets with minimal mortality to the biological control agents. This technology will be used by action agencies to extend the use of the arundo wasp to the far reaches of the Rio Grande river where *Arundo donax* (giant reed) invades.

Optimized aerial application treatments. With rising operational costs, including fuel and chemical inputs, and an increasing concern and awareness of the damaging effects of spray drift away from targeted treatment areas, it is critical that aerial applicators maximize the efficiency of the spray treatments they apply. ARS researchers in College Station, Texas evaluated conventional and innovative application technologies, at varying spray rates and droplet sizes, to determine optimum deposition on the specified target. The work showed that optimum spray deposition within a dense plant canopy can be achieved with significantly larger droplets than those found in small droplet sprays that are highly driftable and that can thus cause damage to non-targeted plants and other negative environmental effects. This accomplishment is important because it provides guidance to the aerial application industry on the proper use of spray treatments that will provide the desired results, while significantly reducing off-target movement of the sprays and the adverse environmental impacts which can result. These results will also help

applicators to address new spray conditions and requirements that may develop due to climate changes in their region.

A monitoring system for detecting herbicide treated fields. The effectiveness of herbicide treatments against pest weeds can vary substantially among different fields, and new methods are needed to accurately map herbicide performance over entire fields or even multiple field complexes. ARS Researchers in College Station, Texas showed that a type of instrument that measures light reflectance off plant surfaces can be effectively used to detect and measure the relative weed killing effects of the herbicide glyphosate. The technique, known as multispectral reflectance, can be adapted for use on either aerial or ground based application equipment. The work is important because it provides a new approach to accurately evaluate herbicide effectiveness under real-world conditions; with the ultimate result that herbicide application protocols can be adjusted to assure maximum effectiveness with a minimum of chemical used.

New grass demography data improves re-vegetation success in rangeland restoration. Degraded rangeland can be difficult to restore because invasive species can often be successful in these systems, while establishment of native species' seedlings is not. Augmentative seeding can help overcome this limitation, however it can be expensive and success rates are variable. ARS scientists in Burns, Oregon, examined the demography of grass species to determine at what life stage seeding failure is most likely, and determined that the most critical period for native species establishment was the transition between germination and emergence. They also demonstrated that important plant traits for establishment for both invasive and native species in low nitrogen soils, such as degraded rangeland, were early germination, root growth at low temperature, and a high specific leaf area. Also, ARS scientists at the Pest Management Research Unit, in Sidney, Montana, demonstrated that the environment in which plants are grown can impact the traits carried over to seed progeny, i.e., drought-stressed plants produce drought-tolerant seeds. The results of these studies will be of value to researchers and land managers in the selection and use of native species lines in the restoration of degraded rangeland.

New biocontrol agents identified for Cape Ivy. Cape ivy is an invasive alien weed from South Africa that smothers native vegetation along the coast of California. ARS scientists in Reno, Nevada, conducted laboratory experiments to evaluate the host (Cape ivy) specificity of a stem-boring moth and a gall-forming fly. Both species are highly specific to Cape ivy, and a petition demonstrating their safety for release was submitted to the USDA APHIS Technical Advisory Group for Biological Control of Weeds. Successful establishment of these agents should reduce the size and abundance of the Cape Ivy weed, reduce control costs, and lead to the reestablishment and improved survival of native vegetation.

Discovery and release of biological control agents of invasive species in Florida. Florida has been hit hard by invasive species because of its subtropical climate and its robust connections to the rest of the world. State and individual water management districts actively attempt to manage select invasive species of plants that threaten natural habitats, using a combination of mechanical, chemical, and biological control, through the development of natural enemies of invasive weeds and insect pests. In many cases, that involves finding biological control agents in the native range of the invasive species. When biological control is successful, it solves the weed or pest problem without the addition of chemicals to the environment and sustains itself through the creation of a natural balance between species. ARS scientists at the Australian Biological Control Laboratory in Brisbane, Australia, the South American Biological Control Laboratory in Buenos Aires, Argentina, and at the Invasive Plant Research Laboratory in Fort Lauderdale, Florida, developed natural enemies against key invasive weed species of Florida. This included the establishment of a gall fly population, *Lophodiplosis trifida*, as a new biological control agent of the melaleuca paperbark tree; the discovery of a rove beetle that attacks skunkvine, an invasive weed that displaces native vegetation; and the completion of years of work to successfully release a new leafhopper biological control agent of waterhyacinth in Florida. These achievements will help preserve the native vegetation and wildlife in Florida at low cost and with minimal management in the future.

Brazilian waterweed management. *Egeria densa* spread in the Sacramento-San Joaquin Delta greatly impacts commercial

navigation as well as potable and irrigation water delivery for over 23 million Californians.. ARS scientists in Reno, Nevada, in collaboration with state and other federal agencies developed an effective herbicide control strategy that can reduce *E. densa* cover by 50 to 75 percent and biomass, by 90 percent ,. This control strategy, when coupled with other methods under development such as biological control, should significantly reduce the impacts of this invasive plant on U.S. waterways.

Weed seed production can be plant density dependent. Yellow starthistle is an important invasive rangeland alien weed in the western United States. Six species of insects that damage flower heads have been introduced for biological control of this weed. However, it is not known whether insects that directly destroy seed are inherently more effective than those that attack other plant parts. ARS scientists in Reno, Nevada showed in field experiments that putting more seed in field plots resulted in more plants if densities were initially low, but that regardless of the number of plants that grew, all the plots produced about the same numbers of seeds one year later. This is because plants at low densities produce more seed than those at high densities, thus compensating for the initially low density of seeds. This demonstrates that population affects on fecundity of invasive plants should be assessed when determining the types of insect natural enemies predators to search for and develop for biological control.

Molecular support for biological control of *Ludwigia hexapetala.* Knowledge of the genetic diversity of invasive plants, such as Ludwigia species, provide valuable insight into the approach needed to manage them. ARS and University of California scientists in Davis, California used amplified fragment length polymorphism (AFLP )to genotype 944 rampets from 32 populations of Ludwigia hexapetala and Ludwigia grandiflora. These samples, which came from five watersheds in California, were evaluated in order to assess the genetic diversity within population of the species. Analyses revealed extremely limited genotypic variation in both species, indicating clonal spread by dispersal of vegetative fragments within and among watersheds, and limited seedling recruitment. These results further indicate that management should focus on limiting vegetative growth and dispersal of fragments, and that the low genetic diversity within

these species improves the likelihood that effective measures of biological control for these invasive species can be developed.

Knowing the invader to improve the success of control. The perennial pepperweed plant invasion in the United States includes more than one species. Plants can be hard to identify to species level using morphology alone,; if the invasive species is misidentified, searches for insects and diseases that can be imported to control the invasion can be done using the wrong plant species or in the wrong region of the world. ARS researchers in Sidney, Montana used DNA data on perennial pepperweed plants from the United States and Asia and found that the invasion in the United States includes two species: *Lepidium latifolium* and *Lepidium affine*. These two species have different ranges in Asia, and perhaps different insect and pathogen agents that attack them. Biological control researchers will now include both of these species in the search for agents that can control the invasion in the United States.

Land-use legacies of dry farming in the Great Basin. Revegetation success is often poor because past disturbances severely altered vegetation and soils. In particular, the impacts of the once common practice of dry farming throughout the Great Basin have not been comprehensively evaluated even though millions of acres of land were cultivated and then abandoned after the dry farming boom from 1910-1920. Because the primary mission of ARS scientists at the Forage and Range Research Laboratory in Logan, Utah is to develop plant materials for revegetation, understanding ecosystem properties, especially vegetation and soils of disturbed regions is critical. In 2010 a detailed evaluation of three different ecosystems was conducted in the Great Basin, using paired sets of historically dry-farmed land and adjacent areas that have never been cultivated. This research identified that land-use legacies of dry farming have striking consequences on vegetation recovery for nearly a century after cultivation has ceased. Land-use legacies in sagebrush ecosystems have direct implications for quantifying ecosystem health, and to make informed predictions about how ecosystems respond to invasions and restoration and re-vegetation efforts. Combined, this novel research will not only assist with developing more appropriate management strategies for reducing the impacts of invasive species on formerly disturbed lands, but provides definitive “soil and

vegetation" indicators to directly assist the selection of plant materials to remedy disturbed soils and the development of improved plant materials.

Immobilizing nitrogen to control plant invasion. Nitrogen pollution has been shown to exacerbate plant invasion in many ecosystems. ARS scientists in Fort Collins, CO, and Logan, UT, in collaboration with Colorado State University scientists, compared management strategies for manipulating nitrogen availability and invasion. Effective management strategies were those that encouraged slow-growing native plants, which can immobilize nitrogen, and reduce the nitrogen available to invasive plants. Such management strategies include ecosystem restoration and, in some instances, grazing or burning. These research findings simplify the relationship between nitrogen pollution and invasive species management, and help land managers determine the feasibility of manipulating nitrogen to inhibit invasion by weeds.

Restoring competitors and natural enemies for long-term control of invasive plants. Managing invasive plant species in rangelands is challenging given the large areas involved and the low economic returns per unit area. Economically sustainable management strategies must therefore be effective over the long term. ARS scientists in Fort Collins, CO, in cooperation with University of Colorado and Colorado State University scientists, determined that biological control and ecosystem restoration can be highly effective regarding long-term control of invasive plants if these management strategies are used together. These findings can be widely used by land managers in determining how to control invasive plants in rangelands with changing environments.

High-resolution aerial surveys detect leafy spurge invasion fronts on wildland. Invasion by weeds is a leading threat to intact native plant communities on wildlands. Detection of weed invasions is often limited to roadside surveys but there is a pressing need to detect invasive weeds occurring in spatially small areas that are often diffuse in arrangement. Timely detection of these localized populations can help in efforts to preserve native plant communities as well as provide land mangers with earlier knowledge of presence of these weeds to increase control efforts. ARS scientists from

Cheyenne, Wyoming, in cooperation with the Bureau of Land Management in Idaho, used a dual-camera aerial survey to determine leafy spurge occurrence and distribution, as well as relate distribution to associated vegetation and control efforts. Simultaneously obtaining images at 2 different resolutions with coordinated fields-of-view optimized leafy spurge detection capacity while retaining plant identification capability. Compared to current ground-based surveys, aerial surveys provide land managers an enhanced capacity for early detection of invasive weeds on wildlands.

## Animal Diseases: : Detection, Identification, Characterization, Prevention, Monitoring, and Control

Vaccines to stop the cattle fever tick. Cattle fever ticks are a world-wide pest of cattle that decimate the economics of the industry by exsanguination of animals and by transmission of key diseases like babesiosis and anaplasmosis. These diseases not only reduce the efficiency of production, they also prevent trade in live animals. The cattle fever tick is a current threat to international food security and an imminent threat to the cattle industry in the southern United States, where the tick was eradicated during a period between 1907 and 1943. Scientists in Kerrville, Texas, collaborating with scientists from EMBRAPA (The Brazilian Agricultural Research Corporation) Brazil, identified two anti-cattle tick vaccine candidates in cattle trials. These candidates had been prioritized in a prior Agricultural Research Service project through bioinformatic and molecular biological approaches. In the cattle trials conducted in Brazil, the candidates outperformed the recombinant Bm86 Campo Grande antigen, which is an antigen similar to that used in the only current commercially available anti-tick vaccine. An invention disclosure was filed and cattle trials are scheduled to evaluate various parameters in the vaccination protocol to optimize efficacy. The worldwide use of a consistently effective anti-tick vaccine in cattle would reduce production costs associated with tick treatment and contribute to the maintenance of the eradication of cattle fever tick in the United States.

Developed a statistical model for predicting influenza virus transmission. Modeling is an important component of predicting the spread of diseases. Often it is possible to develop and adapt systems

for modeling human diseases to those that affect livestock animals. ARS researchers at Beltsville, MD have developed a model for the human H1N1 influenza virus transmission in collaboration with scientists at Northeast Agricultural University, Harbin, China. The availability of extensive human data provides support that such a model will be adaptable to swine viral diseases. Swine H1N1 cases in Mainland China from May 13 to July 22, 2009 were analyzed with this novel mathematical model to better predict future infection trends. Model efficiency was evaluated by numerous methods all of which demonstrated a high predictive capacity. This algorithm was a substantial improvement over the most commonly used algorithm, the grey theory, which had no predictive value for this dataset. The success of this equation suggests its potential adaptability to other animal infections.

PRRS virus non-structural proteins exhibit increased host pathology. Porcine reproductive and respiratory syndrome (PRRS) is a serious swine disease that appeared suddenly in the midwestern United States and central Europe approximately 14 years ago; the disease has now spread worldwide. ARS researchers at Beltsville, MD, partnered with South Dakota State University (SDSU), and Kansas State University scientists to probe the function of the Porcine Reproductive and Respiratory Syndrome virus (PRRSV) non-structural proteins (nsps) in viral pathogenesis and host immunity. Recently, virulent Porcine High Fever Disease associated with PRRS virus variants, with deletions in the PRRSV nsp genes, were reported in China and linked to the high morbidity and mortality observed. At SDSU a complete PRRSV infectious clone was produced; it was then genetically altered at specific nsp positions, termed epitopes because of their role in immune responses, and new PRRSV infectious clones characterized. One nsp mutant, PRRSV-deletion ES3, was actually more infectious and caused higher pathology than the parental virus. When tested this ES3 clone stimulated less interleukin-1 beta (IL-1b) and tumor necrosis factor (TNF) indicating that the innate immune responses were altered. This ES3 deletion mutant caused higher pathology both in vitro and in vivo when compared to the parental virus or other PRRSV deletion mutants and provides valuable information for planning improved PRRS control strategies, including improved vaccines. These plans will be essential to help prevent the spread of Porcine High Fever Disease from China.

Developed sensitive multiplex Luminex bead assay for swine cytokines. ARS researchers at Beltsville, MD, partnered with South Dakota State University (SDSU) to develop a Luminex bead based multiplex assay, or Fluorescent Microsphere Assay (FMIA), to measure 8 swine cytokines simultaneously in pig serum. The assay detects cytokines that regulate several aspects of the swine immune system, the early innate [interleukin-1beta (IL-1b), IL-8, interferon-alpha (IFNa), TNFa]; the regulatory (IL-10), the T helper 1 (IL-12, IFNg) and T helper 2 (IL-4) cytokines. The advantage of a 'multiplexed' assay is that multiple cytokines can be evaluated at one time, only a small volume of each sample is needed and there is a broad dynamic range of marker sensitivity. The assay was tested on sera collected from a porcine reproductive and respiratory syndrome virus (PRRSV) vaccine/challenge study. Multiple serum cytokines were altered by PRRSV vaccination and challenge; but only significantly elevated levels of IL-12 were observed in the killed vaccine/adjuvant group. However, this significant increase in serum IL-12 correlated with lack of protection against PRRSV viremia indicating the unique role of this cytokine in pigs. This assay provides a major new tool to define protective immune responses in PRRS and other respiratory diseases in pigs and importantly for designing more efficacious vaccines for swine pathogens.

Characterized early events of Foot-and-Mouth Disease Virus pathogenesis in cattle. Understanding the basic mechanisms of viral pathogenesis, including the viral and host determinants of virulence is pivotal to developing effective control and eradication tools. ARS researchers at PIADC, Greenport, NY have combined veterinary pathology with molecular biology and bioinformatics approaches in order to better understand the early (previremic) FMDV – host interaction after aerosol infection in cattle. FMDV was most consistently localized to nasopharyngeal tissues, thereby indicating this region as the most important site of primary viral replication. The earliest site of microscopic localization of FMDV antigens was cytokeratin-positive pharyngeal epithelial cells. Onset of viremia coincided with marked increase of viral loads in pulmonary tissues and with substantial decrease of viral detection in nasopharyngeal tissues. These data indicate that subsequent to aerogenous exposure to FMDV, the temporally defined critical pathogenesis events involve

(1) primary replication in epithelial cells of the pharyngeal mucosa-associated lymphoid tissue crypts and (2) subsequent widespread replication in pneumocytes in the lungs, which coincides with (3) the establishment of sustained viremia. A mutant FMDV with impaired initiation of translation replicated in the pharynx but did not invade the lung, had no detectable viremia and did not cause clinical signs confirming that lung invasion is necessary for viral generalization and disease. The application of functional genomics to the understanding of the FMD virus – host interaction in a relevant animal inoculation model uncovered novel pathogenesis mechanisms and deepened our understanding of this relevant animal disease.

Showed inhibition of innate immune responses to early infection with Foot-and-Mouth Disease Virus (FMDV). Analysis of peripheral blood cells from swine by ARS researchers at PIADC, Greenport, NY showed a cell population that is the primary source of early anti-viral response. These cells, termed "the natural killer" (NK) cells, produce the antiviral protein interferon gamma (IFN gamma). Analysis of these NK cells isolated from animals infected with FMDV show inhibition of the IFN gamma production, compromising the anti-viral response in infected pigs. This correlates with the interruption of similar responses by other cells, specifically dendritic cells, shown previously in infected animals. These data show that infection with FMV renders important, early responding cell populations less active, allowing the virus to spread throughout the animal and between animals.

Identified a key factor for Foot-and-Mouth Disease Virus replication. Work by ARS researchers at PIADC, Greenport, NY in this period has demonstrated for the first time the involvement of the RNA helicase A (RHS), a nuclear cell protein, in foot-and-mouth disease virus replication. This protein, which in normally located in the nucleus was shown to relocate to the cell cytoplasm to enhanced virus replication. The molecular mechanisms of RHA function was shown to relate to its ability to interact with both, virus proteins and with the viral genome. This information uncovered a novel host factor and aid in our understanding of the mechanism of virus-host interaction during infection.

Determined host-cell gene expression in response to FMDV viral

infection. ARS researchers at PIADC, Greenport, NY using microarray technology the gene expression profile of bovine cells infected with wild type (WT) FMDV or an attenuated virus lacking the leader protein (leaderless FMDV) was determined. Thirty nine out of approximately 22,000 bovine genes were selectively up-regulated by 2 fold or more in leaderless versus WT FMDV infected cells. Many of these genes were involved in regulation of the innate immune responses. A novel host response gene was identified in response to FMDV infection and is currently being explored as a biotherapeutics candidate to prevent FMDV infection.

Testing of potential live-attenuated Foot and Mouth Disease (FMD) vaccine candidates in swine. ARS researchers at PIADC, Greenport, NY have developed improved vaccine platforms and delivery systems to control FMD. Using bioinformatic tools protein domains have been identified in the FMDV leader coding region. Mutation of specific amino acid residues within this region resulted in viruses that grow to high titers in tissue culture but showed reduced virulence in swine and cattle eliciting a strong neutralizing antibody response. Preliminary studies in swine demonstrated that live attenuated vaccine candidate can protect against virulent virus challenge. This attenuated FMDV can also be also as an inactivated vaccine platform.

Testing of a new biotherapeutic agent in cattle. A novel biotherapeutic reagent for foot-and-mouth disease (FMD) was identified by ARS researchers at PIADC, Greenport, NY using microarray analysis of cells infected with wild type and attenuated FMDV. This novel biotherapeutic was cloned using an Ad5 expression vector. Protein expressed from this construct induces antiviral activity against FMDV in vitro and in vivo. Inoculation of cattle with Ad5 expressing the biotherapeutics induces the expression of several IFN induced genes and delays FMD infection.

Enhanced efficacy of Ad5-pIFNalpha vector in swine. Enhanced the efficacy of Ad5-pIFNalpha vector against FMDV challenge in swine. ARS researchers at PIADC, Greenport, NY have demonstrated that delivery of the vector at 4 sites in the neck subcutaneously reduced the protective dose as compared to intramuscular vector delivery. Performed a number of assays to demonstrate that Ad5-pIFNalpha

could sterilely protect swine against FMDV challenge.

Improvement of vaccine virus growth characteristics. ARS researchers at PIADC, Greenport, NY have initiated a study to identify determinants of foot-and-mouth disease virus (FMDV) tissue culture adaptation that could be used to enhance the properties of vaccine candidates derived using the recombinant DNA technology. This project is a component of a multi-national collaborative effort to enhance the stability and adaptation of FMDV candidates for vaccine production. We have amplified a selected type A FMDV and performed genetic and phenotypic characterization. Initial characterization of eield viruses included determination of viral capsid sequences and the selection of mutation to be engineered in these viruses to ehnance virus growth. Primers and other reagents needed to molecularly clone these viruses were attained.

FMDV leader deleted virus as a vaccine candidate and further development of a companion diagnostic test. Two experimental FMD vaccines carrying either one or two negative markers protected animals from challenge with the parental virus. The attenuated characteristics and inability to spread to in contact animals was also demonstrated by ARS researchers at PIADC, Greenport, NY for these negative marker vaccine viruses in susceptible animals, including cattle and pigs. A companion ELISA test was adapted to provide a way to distinguish infected from vaccinated animals.

Identification of determinants of foot-and-mouth disease virus adaptation to cell culture. ARS researchers at PIADC, Greenport, NY in collaboration with OVI-Onderstepoort on determinant for tissue culture adaptation of SAT FMDV have led to the identification of unique Heparan Sulphate Proteoglycans-binding sites (amino acid residues) located on the outer capsid proteins of SAT1 and SAT2 FMDV. The sites are exposed on the surface of the virion and are structurally accessible for binding to the alternative HSPG receptors.

Reactive cytotoxic t lymphocytes against homologous and heterologous avian influenza subtypes. Numerous reports have implicated a role of genetic resistance to bacterial infection and viral diseases. However, little is known about the role of genetics of chickens in generating protective immunity following avian influenza

(AI) infection. In these studies, genetically-defined chickens were infected with a recent H9N2 AI isolate and thymus (T)-derived lymphocytes were analyzed for cross reactivity against different AI viruses. Results indicate cells isolated from H9N2-infected chickens displayed lysis of lung cells infected with many AI isolates. Removal of a specific portion of these cells, the CD8+ population, removed the specific immunological cross reactivity. Taken together, these studies provide insight into the cross reactive nature of avian T lymphocytes against AI viruses.

Efficacy of recombinant herpesvirus-of-turkeys vaccine against mexican-lineage avian influenza H5N2. In Mexico, outbreaks due to low pathogenic (LP) avian influenza virus (AIV) H5N2 strains started in 1993-1994 and several highly pathogenic (HP) strains emerged in 1994-1995. Although the HP strains were contained and have not been reported since 1996, LP strains remain endemic in Mexico despite an extensive vaccination program. The objective of this study is to evaluate the efficacy of turkey herpesvirus (HVT) vectored AIV vaccines (H5 subtype) against challenge with Mexican lineage H5N2 AIV strains. The avian influenza (AI) HA gene from two different H5 AIV was cloned into a HVT vaccine. Following challenge with a lethal dose of H5N2 HPAI, most all birds receiving either of the HVT-AI vaccine survived. These results indicate this type of recombinant vaccine can be used as an aid during AI eradication efforts.

Development of enzyme linked immunospot assay (elispot) to detect avian influenza specific antibody-secreting B cells in chickens. Vaccines remain a cost-effective means to protect animals from infectious disease by establishing immunity following application. To evaluate next generation vaccines, new methodologies are needed to expand analysis of antibody producing cells in the host after vaccination. A novel ELISPOT method was developed which allows detection and enumeration of antibody producing cells from chickens against avian influenza (AI). Inactivated and labeled AI virus was incubated with lymphocytes from AI-vaccinated chickens and the number of cells producing antibodies against AI determined. With this method one can identify and enumerate both the total number of antibody secreting cells and those secreting antibodies to AI in a highly sensitive manner and at the cellular level. In this way, AI vaccines and adjuvants inducing higher numbers of antigen-specific

antibody secreting cells may be developed and compared to currently available vaccines.

Differential growth characteristics of avian influenza viruses in primary cell culture. Low pathogenic avian influenza (LPAI) viruses cause varying pathogenicities when inoculated into chickens. Infection with some LPAI isolates results in no overt signs of clinical disease, while others cause respiratory distress, weight loss and diarrhea. In these studies the growth characteristics of four different LPAI isolates was compared in primary chicken cell cultures. Results indicate two H5 isolates grew poorly in chicken embryo liver (CEL) cells, compared to either a H7N2 or H9N2 LPAI isolate. In contrast, all four LPAI viruses grew to higher titers in chicken embryo kidney (CEK) cells. The presence of trypsin in the growth media increased the titers of the LPAI viruses in the CEL cells, but had no effect on titer in the CEK cells. Taken together these results shed insight into the varying growth characteristics displayed by LPAI viruses of differing pathogenicities.

Intranasal administration of alpha intreferon reduces morbidity associated with low pathogenic avian influenza infection. Type I interferons, including interferon alpha (IFN-alpha), are expressed rapidly after viral infection, and represent a first line of defense against avian influenza. Following infection of chickens with avian influenza virus (AIV), transcription of IFN-alpha is quickly up regulated along with a myriad of other immune-related genes. In these studies, we assessed the protective potential of IFN-alpha applied to birds prior to exposure to low pathogenic AIV. Intranasal application with IFN-alpha prior to and during active AIV infection reduced clinical signs of disease, including weight loss and fever, compared to phosphate-buffered saline (PBS) treated controls. In addition, the incidence of viral shedding and viral titers from oral swabs was significantly reduced in IFN-alpha treated birds. Taken together, these studies show that IFN-alpha can protect chickens from disease associated with low pathogenic AIV and reduce the risk of transmission through decreased shedding.

Adaptive transfer of lymphocytes from avian influenza infected chickens protects from overt clinical signs of disease following infection with H9N2 low pathogenic avian influenza. Immunity

against avian influenza (AI) is largely based on the induction of neutralizing antibodies produced against the hemagglutinin, although host lymphocytes have been reported as critical for clearance of virus from infected cells. In these studies, chickens were infected with a recent H9N2 AI isolate, and lymphocytes from those birds adaptively transferred to immunologically naïve birds. The birds receiving the lymphocytes produced against AI were then challenged with the H9N2 virus. Results indicate lymphocytes from infected birds could protect naïve birds from overt clinical signs of disease. In contrast, control birds had significant decreases in body weight and higher body temperatures following H9N2 infection. These studies demonstrate the protective nature of cell mediated immunity of chickens against AI viruses.

Studying the effect of NS1 gene exchange on the pathogenicity of H5N1 highly pathogenic avian influenza viruses in ducks. Until 2002, H5N1 highly pathogenic avian influenza (HPAI) viruses caused only mild respiratory infections in ducks. Since then, new viruses have emerged that cause clinical disease and high mortality in ducks and other waterfowl. However, there is no clear explanation of why the pathogenicity of some H5N1 HPAI viruses has increased. The NS1 influenza virus protein is known to suppress immune responses in virus-infected hosts, consequently affecting the virus pathogenicity. In order to determine if the NS1 protein contributes to the increased virulence in ducks, single gene reassortant viruses were generated. The NS1 gene from a virus that produces mild disease in ducks and from a very virulent virus for ducks were exchanged for the NS1 of a moderately pathogenic virus in ducks. Exchanging the NS1 gene had minimal effect on the pathogenicity of the virus, and suggests that other viral genes, or combination of genes, are most likely contributing to the increased virulence of H5N1 HPAI viruses in ducks.

Genetic and aminoacid comparisons of pandemic H1N1 to U.S. H1N1 avian influenza vaccine isolates. In 2009, a pandemic influenza A H1N1 (pH1N1) virus was isolated in swine in Canada in June, and later in turkey breeders in Chile, Canada, and the U.S. The pH1N1 virus consists of gene segments of avian, human and swine influenza origin and raises the potential for infection in poultry following exposure to infected humans or swine. In these studies, the

relatedness of the hemagglutinin (HA) gene segments from the pH1N1 to U.S. H1N1 AI isolates used as inactivated vaccines in commercial turkeys was determined. Genetic analysis indicates U.S. H1N1 AI vaccine isolates contained between 76- 92 % nucleotide sequence similarity to the pH1N1 virus. However, comparison of amino acids found at antigenic sites of the hemagglutinin (HA) protein indicate major differences were found between pH1N1 and the U.S. H1N1 vaccine isolates. Taken together these results suggest limited cross reactivity between U.S. H1N1 vaccines and the pH1N1 virus. Current vaccines used in turkey breeders against circulating H1N1 viruses should be updated and tested to ensure adequate protection for field exposure.

Genetic characterization of new exotic viruses. Virulent Newcastle disease viruses (NDV) are found in most countries of the world and the United States has strict rules to prevent their entry. However, it is important for us to monitor and characterize viruses that are a potential threat to the U.S. poultry industry. NDV obtained recently from Mexico, Indonesia, Malaysia, Venezuela, Pakistan, Vietbam, Bekuze, Dominican Republic, South Africa and from wildbirds in the U.S. have been genetically sequenced and characterized phylogenetically. The sequence data allows the current diagnostic tests for NDV to be kept current to ensure that the circulating viruses can be properly diagnosed.

Level of immunity against Newcastle disease viruses (NDV) affects transmission after a virulent challenge. Current commercial Newcastle disease (ND) vaccines, when given correctly, protect birds from dying or getting sick after infection with virulent viruses, but they do not protect vaccinated birds from being infected and from shedding viruses to other birds. We assessed the relationship between the amount of antibodies produced by vaccinated animals and the capacity to transmit challenge viruses. Birds vaccinated with live ND vaccines were challenged at different times post vaccination to test vaccine efficacy to prevent transmission. Vaccinated birds that were challenged before 21 days post vaccination were able to transmit viruses to vaccinated and non-vaccinated birds and shed significantly more viruses in oral secretions than birds challenged after 21 days. This information suggest that an earlier onset of immunity may help protect against transmission and spread of the

disease.

Pathogenic characterization of exotic viruses. Newcastle disease viruses (NDV) with different genomes behave differently in poultry and the clinical signs and lesions seen can vary greatly. Pathogenesis experiments are needed to ensure that we can identify the disease if it entered into our poultry. We initiated the pathotyping of new viral isolates that have the potential to be introduced in the US. Pathogenic characterization of viruses from China, Peru, Dominican Republic, Belize, Vietnam and US waterfowl has been done in chickens. The data allows for the identification of the disease signs which are crucial to quickly preventing the spread NDV.

Laboratory reproduction of "egg drops" seen in the field. A vaccine experiment (Objective 2), was performed to develop an animal model that mimics the decrease in egg production seen in well vaccinated animals in Mexico, Korea and South Africa. In addition, an initial pathology study was performed to evaluate multiple sections of the reproductive tract of laying hens for microscopic lesions and for evidence of Newcastle disease virus (NDV) infection. Lesions were found in the hens that did not receive an additional vaccine boost eleven days after infection. It has been documented since the 1950s that egg production, even in well vaccinate birds, is a problem when flocks are infected with virulent NDV. This initial test will be extended to continue to understand the disease process and how the reproductive tract is affected.

Characterization of a new avian paramyxovirus isolated from penguins. There are many avian paramyxoviruses that can infect Newcastle disease viruses (NDV) but never get characterized because there are not defined systems to do so. As a result of a surveillance program in wild birds a hemagglutinating virus that was not recognized with current diagnostic test was identified. Characterization of new types of paramyxoviruses is important from the diagnostic perspective, as they may be confused with Newcastle disease viruses. The virus was sequenced using a random approach and the serology characterized. It was determined that the viruses corresponded to a new serotype (serotype 10).

Sample processing for avian influenza virus diagnostic test improved to increase test sensitivity. Cloacal swab samples from poultry and wild birds are a common sample type for avian influenza virus detection, but the fecal material in these samples often contains substances that will inhibit the diagnostic test. ARS researchers in Athens, GA developed a new method of processing the specimens which effectively washes away the inhibitors. This results in a substantial increase in test sensitivity and therefore results in more accurate detection of avian influenza virus from poultry and wild birds.

Turkey susceptibility to and transmission of influenza viruses. Influenza viruses from numerous species of wild birds and domestic poultry were tested for their ability to infect domestic turkeys, chickens and ducks. ARS scientists in Athens, GA helped demonstrate that turkeys could be most easily infected in comparison to chickens and ducks as they were susceptible to the lowest doses. Turkeys were also susceptible to swine influenza which is frequently observed in the field. These data show that turkeys may be more susceptible for influenza from numerous avian and mammalian species, thus emphasizing the importance of disease control and that turkeys are highly susceptible hosts that have the potential to mediate infection between species.

Development of an oral vaccine for avian influenza using yeast cells. Traditional vaccination methods for high pathogenicity avian influenza viruses require costly and time-consuming injection of individual birds, often multiple times, in order to provide adequate protection. The use of yeast as an expression system for influenza proteins can potentially be inexpensive and can be given in the feed. Yeast also provides high quality nutrition when added to the feed. ARS scientists in Athens, GA, successfully expressed the hemagglutinin protein from a subtype H5N1 highly pathogenic avian influenza virus (the protective protein in flu vaccines) on the surface of the yeast strain Pichia pastoris. Functionally, the protein retained its function in the yeast and oral vaccination of chickens produced antibodies which could block influenza infection. This study represents the first step in the development of a yeast-based vaccine for poultry for highly pathogenic strains of avian influenza virus that can be administered in feed.

The 2009 pandemic H1N1 influenza virus does not easily infect young poultry. When the pandemic H1N1 influenza virus emerged in the spring of 2009 it was unknown whether the virus could cause disease in poultry or whether poultry could act as reservoir for the virus. ARS scientists in Athens, GA determined that young chickens and turkeys could not be infected by respiratory route, however low levels of virus could be recovered from quail. This demonstrated that the virus is poorly adapted to poultry, therefore poultry would likely not serve as a reservoir and the virus has a minimal disease potential for young poultry.

Adult turkey hens are susceptible to 2009 pandemic H1N1 virus by reproductive tract insemination. In contrast to information about infection of young poultry with the human pandemic H1N1 virus, turkey breeder flocks world-wide became infected with the H1N1 pandemic virus, presumably from infected humans as there was no epidemiological data for other birds as a source of infection. In order to understand how this occurred ARS scientists from Athens, GA exposed adult breeder age turkeys to the pandemic H1N1 by the respiratory route and through the reproductive tract by artificial insemination procedures. Only the turkeys that were exposed by artificial insemination methods were infected. These results suggest that the turkey breeders could have been infected by infected insemination crews and that biosecurity practices for artificial insemination, which is universally practiced by the turkey industry, need to be modified.

Superior H5 avian influenza virus strains were identified for vaccines for Central American poultry. Vaccination is widely used to control avian influenza virus in Latin America, however the virus continually changes so that it can evade the immune system, therefore vaccines that were produced with a virus from 1994 were no longer adequately effective. ARS scientists in Athens, GA tested the old vaccine and new strains for their ability to protect chickens against recent strains circulating in Central America. New strains that provided superior protection were identified. Vaccine manufacturers can utilize these new strains of virus to make improved vaccines for the control of avian influenza virus. Improved control of avian influenza virus in Central America reduces the risk of virus infecting US poultry.

The pathogenesis of the 2009 pandemic H1N1 virus was characterized in mice and was found to be milder than high pathogenicity avian influenza virus. The mouse is a widely used model system for studying influenza virus pathogenesis and has been used to look at numerous mammalian and avian influenza viruses, and thus contributes invaluable information about the transmission and biology of the virus in different species. ARS scientists from Athens, GA compared the pandemic H1N1 with several other influenza viruses including the Asian high pathogenicity H5N1 virus and found that the pandemic virus caused relatively mild damage in the lungs. Mouse models have been crucial for evaluating and understanding influenza strains from many species and can help assess their pandemic potential, evaluate vaccines and identify species specific characteristics.

The ecology and dissemination of high pathogenicity avian influenza viruses in Pakistan was characterized by completing genetic analysis on isolates from 1995 to 2004. High pathogenicity avian influenza viruses of the H7N3 subtypes have persisted in poultry in Pakistan despite vaccination and bio-security programs, therefore it has been unclear whether these are new introductions of virus or whether the virus is maintained within Pakistan with occasional transmission to poultry. ARS scientists from Athens, GA produced full genome sequence of these viruses and discovered that the viruses in Pakistan are distinct from other avian influenza viruses indicating that the virus is maintained in a reservoir within the country. They also found that the virus was genetically mixing with other influenza subtypes that were infecting poultry. Now that the source of the virus has been identified as being within the country and not new introductions from other regions, control programs can be developed to focus on local biosecurity. Additionally, this provides broader information on the ecology and dissemination of high pathogenicity avian influenza viruses which could be introduced into US poultry, which enhances US control programs.

A South American avian influenza virus (AIV) isolate with genes related to both the 2002 high pathogenicity virus in Chilean poultry and North American wild bird viruses was discovered. Prior to an outbreak of high pathogenicity AIV in poultry in Chile in 2002, no AIV had been reported in South America and little was known about AIV

in wild birds in South America. However, ARS scientists from Athens, GA discovered an AIV in wild bird specimens collected in Bolivia in 2001. Genetic analysis of the virus revealed that some genes were related to the virsues from 2002 in Chile and some genes were related to wild bird viruses from North America. The source of the virus was a cinnamon teal (Anas cyanoptera), which is a non-migratory duck, indicating that AIV is found in South American wild birds.

Age at infection affects the pathogenicity of Asian highly pathogenic avian influenza H5N1 viruses (AIV) in ducks. A unique trait of the Asian H5N1 high pathogenicity influenza viruses is that some strains have developed the ability to cause disease in waterfowl, such as ducks. ARS scientists from Athens, GA evaluated the ability of several of these strains to affect 2 and 5 week old ducks. Although the severity of disease and mortality was somewhat dependent on the strain used, the older ducks were much more likely to survive infection and experienced less severe disease. It was also found that disease correlates with the level of viral replication in tissues. This reveals a critical aspect of the Asian H5N1 AIV biology and can aid control programs by focusing on prevention of infection in young ducks.

Commercial vaccines have variable efficacy for protecting chickens and ducks against H5N1 highly pathogenic avian influenza (HPAI) viruses from Vietnam. Vaccination is the primary control method that has been employed for avian influenza viruses in Vietnam and there are numerous commercial vaccines, but little data is available about how well they work. ARS scientists in Athens, GA evaluated three commercial vaccines. The vaccines provided different levels of protection in chickens and ducks following infection with HPAI H5N1 and some were protected from mortality, but viral shedding occurred for at least 5 days post challenge depending on the vaccine, species and challenge virus used. Although the vaccines tested were effective in protecting against disease and mortality, updated and more efficacious vaccines are needed to maintain optimal protection.

The optimal detection methods for avian influenza virus (AIV) from wild birds depends on the prevalence of virus. Surveillance for AIV in wild birds is conducted worldwide and the detection methods

employed depend on the resources of the labs conducting the surveillance. In order to determine the optimal methods for AIV detection in specimens from wild birds ARS scientists from Athens, GA compared numerous methods; cell culture, chicken eggs and real-time reverse transcriptase-polymerase chain reaction (RT-PCR) (which detects the genetic material of the virus). When cost and virus recovery were taken into account it was shown that the most cost effective method depended on whether most of the specimens were from infected birds or not; the cheapest method, real-time RT-PCR was best when infection rates were low, but virus isolation in chickens' eggs was better if infection rates were high.

H6N2 low pathogenicity viruses from poultry in CA and NY are adapted to chickens and not ducks making detection of infection difficult. H6N2 is a common subtype of low pathogenicity avian influenza virus seen in poultry and wild birds in North America. ARS scientists from Athens, GA evaluated several isolates from chickens and turkeys in CA and NY for their ability to cause disease in chickens and ducks. The viruses infected chickens more easily than ducks, but caused only minimal disease in either species. This indicates that these viruses have been circulating in chickens long enough to adapt to them and become less adapted to ducks. Since the disease was so mild and low amounts of virus was shed, yet were adequate to spread among chickens, there is a danger that these virsues could circulate without being easily detected.

Wild birds can disseminate avian influenza virus (AIV) widely throughout Asia. Samples were collected from wild birds throughout Mongolia from 2005-2007 for AIV testing. Since Mongolia has little poultry production any virus isolates were assumed to be disseminated by wild birds only. In addition to the Asian H5N1 high pathogenicity virus strain, ARS scientists in Athens, GA isolated 10 low pathogenicity AIVs from numerous species, and influenza was detected in specimens from two species for the first time (Luscinia svecica and Calandrella cheleensis). Genetic analysis revealed that there was variation among the viruses indicating that each year new AIVs are introduced into wild bird populations in Asia where they can mix with older viruses.

Improvement of a nucleotide polymorphism-based typing system for Escherichia coli O157:H7. Cattle are a reservoir of the recently emerged human pathogen E. coli O157:H7 and harbor distinct subtypes that do not all associate with human disease. A set of 178 nucleotide polymorphisms was developed in 2009 by ARS scientists at Clay Center, NE, that showed great utility in classifying E. coli O157:H7 genetic subtypes of cattle and/or human origin. To achieve higher resolution with this system, the scientists developed and applied a set of 720 nucleotide polymorphisms to 420 E. coli O157:H7 strains. This set effectively tags E. coli O157:H7 genetic diversity associated with humans and cattle, and an evolutionary analysis of the diversity indicates that cattle harbor subtypes that have evolved away from an association with human disease. Consequently, the nucleotide polymorphism set depicts E. coli O157:H7 genetic diversity and shows how strains are evolutionarily related with high resolution. These qualities define it as a preferential method for detecting and distinguishing E. coli O157:H7 genetic subtypes involved in epidemiological investigations.

Introduction of exotic strains of bluetongue virus (BTV) to the US is a constant threat to US livestock. Many exotic strains have been identified in the past few years. Of particular concern for introduction is the European BTV-8 strain (EU-BTV-8). This virus strain has caused devastating disease all across Northern Europe where the disease had not been seen previously. To determine the susceptibility of North American white-tailed deer to EU-BTV-8 infection, an experimental infection study was conducted by ABADRU scientists in collaboration with scientists from Colorado State University and APHIS, National Wildlife Research Center in Fort Collins, CO. Tissue and blood samples were tested for the presence of virus and antibody. Infection resulted in significant clinical disease with peak viremia at 7-10 days. Virus-positive tissues included kidney and liver. This study demonstrates that our North American deer would be highly susceptible to an outbreak of this European BTV-8 were it to be introduced into the US.

Development and preliminary validation of a Rift Valley Fever (RVF) immunological assay. Rift Valley Fever is a deadly Sub-Saharan African disease of livestock and humans. There are limited diagnostic assays available should RVF be introduced into the United

States. Diagnostic reagents that do not pose health risks to the producer and user based on recombinantly expressed target proteins were developed. These reagents were incorporated into assays by ARS scientists in Manhattan, KS, that detected specific antibodies in sheep experimentally infected with RVF MP12 or wild-type virus. Testing of sequential serum samples from experimentally infected sheep obtained from South Africa and the Canadian Food Inspection Agency showed a rise in specific antibody to RVF. These assays provide a safer assay to produce and use thus providing a tool to be used for early detection through distribution to the National Animal Health Laboratory Network.

Characterization of Nor98 in Canadian sheep. Identification and characterization of novel scrapie strains, particularly those in sheep considered resistant to classical scrapie, are key components in the US scrapie eradication effort. In association with the USDA Animal Plant Health Inspection Service's National Veterinary Service Laboratory, ARS scientists in Pullman, WA have previously characterized the Nor98 form of scrapie in US sheep. These scientists have now provided assistance to the Canadian Food Inspection Agency in their characterization of the disorder in Canadian sheep. The disease profiles of all samples were consistent with Nor98 scrapie, a strain that may not be contagious and may be a spontaneous degenerative condition of older sheep. This finding supports the current genetically based control program for scrapie in the US and Canada.

Prion genotypes in US and Canadian Sheep are similar. Selection for genetically resistant animals is a foundation of the current scrapie control programs in the US and Canada. In association with the Canadian Food Inspection Agency, ARS researchers in Pullman, WA described the prion genotypes of Canadian sheep diagnosed with scrapie between 1998 and 2008. The susceptible genotypes used in the US scrapie control program were similar to those found in the Canadian study. This finding allows the US and Canada to continue to harmonize their scrapie eradication programs.

Prion genotypes of US goats are diverse. The role of prion genetics in sheep scrapie is now well described and selection for genetically resistant animals is a key component of the eradication program.

However, the role of prion genetics in goat scrapie is not well understood. More importantly, basic information on the extent of prion gene variation in US goat breeds has been lacking. ARS researchers in Pullman, WA have described the wide variation in gene sequence among several economically important breeds of US goats. This study provides the basis for further examination of the role of prion genotypes in scrapie prevention.

Disease progression in Rocky Mountain elk with chronic wasting disease. Chronic wasting disease (CWD) is the prion disorder of deer and elk. Detailed analyses of the pathways through which the infectious protein moves in the tissues of infected elk are the basis for improved diagnostic testing and control program. In collaboration with Colorado State University, ARS scientists from Pullman, WA, have demonstrated a detailed examination of diseased elk and the pathways through which the agent appears to spread in the brain and eyes. This finding supports the diagnostic testing methods developed by ARS and currently conducted to monitor herds of elk for this fatal disease.

Chronic wasting disease in non-native species. Chronic wasting disease (CWD) is reported in white tailed deer, black tailed deer, mule deer, Rocky Mountain elk, and Shira's moose in the US and Canada. The entire host range of this disorder is not known and facilities housing non-native deer or elk species remain at potential risk of infection if those species are susceptible. In this study, ARS researchers in Pullman, WA provided collaborative assistance to the Canadian Food Inspection Agency in a study examining the effect of experimental infection of red deer with CWD. The study demonstrated the susceptibility of this species to disease, the potential for diagnosis early in the disease, and the role of naturally occurring variation in the prion gene on disease susceptibility. The study demonstrated that diagnostic methods developed by ARS are suitable for use in red deer.

Molecular kinship studies in white tailed deer with chronic wasting disease. Chronic wasting disease (CWD) in white tailed deer can result in very high infection rates with very little evidence of clinical disease in infected animals, even late in disease. The mechanisms by which disease spreads in captive herds are not known. ARS

researchers in Pullman, WA collaborated with scientists at the Veterinary Genetics Laboratory wildlife disease unit in Davis, CA to develop a panel of genetic markers that identify the family structure in herds of deer. Using this panel, epidemiologic studies to examine the transmission of the disease among family members in a herd are now possible.

The role of tissue mineral levels in prion disease in Rocky Mountain elk. Several preliminary studies have suggested an association between the levels of certain dietary minerals in the tissues of elk with chronic wasting disease infection. In collaboration with Colorado State University, the National Park Service, and the USDA Animal Plant Health Inspection Service, ARS researchers in Pullman, WA have shown the increased risk of chronic wasting disease in elk with decreased magnesium and increased manganese levels in brain tissue. This study provides important information on factors affecting disease in the natural host.

Early detection of prion diseases such as sheep scrapie requires concentration of the prion proteins that serve as disease markers from dilute biological samples. Researchers at the Foodborne Contaminants Research Unit in Albany, California, in collaboration with the University of California San Francisco, developed a new method for concentrating prion proteins from animal tissues. We found this method results in a significant increase in prion concentration, allowing more sensitive prion detection. This year we filed a patent application and published a manuscript on this new method, showing how it provides more sensitive and early detection of disease in infected animals.

New strain of mouse for anti-prion antibody production. Production of antibodies for detection of prion proteins associated with diseases such as sheep scrapie is limited by the resistance of normal mice to making an immune response against their own proteins. Researchers at the Foodborne Contaminants Research Unit in Albany, California, in collaboration with our partners at the University of California San Francisco, have made a new strain of mouse that lacks the prion protein. Unlike normal mice, the new mice are highly sensitive to immunization with prions. This year we filed a patent application and published a manuscript showing the use of these

mice in making new antibodies that can bind and detect prions. Such antibodies may be used for more sensitive and early detection of disease in infected animals.

New method for identification of antibodies that detect prion proteins. Making new monoclonal antibodies for detection of prion proteins in diseases such as sheep scrapie requires selection of the best performing cells from among thousands of candidates. Researchers at the Foodborne Contaminants Research Unit in Albany, California have developed a fast and sensitive method to identify such cells, which are taken from mice that have been immunized to produce antibodies that strongly bind to prion protein. This method was used to identify improved antibodies which are now available for use in detection of disease. Furthermore, the new screening method may be used by other scientists for research to make additional new antibodies for prions.

New antibodies for detection of prion disease. Sensitive detection of prion proteins for early diagnosis of disease requires antibodies that are capable of strong binding to prions. Researchers at the Foodborne Contaminants Research Unit in Albany, California, in collaboration with our partners at the University of California San Francisco, generated eight new antibodies that detect prion disease in many different animals. We have published a manuscript that describes these anti-prion antibodies and shows their value in improving detection of prions. Improved detection methods will help in herd management and control of the spread of prion diseases.

Inoculation of domestic and European bovine spongiform encephalopathy (BSE) isolates into cattle. To date, no side-by-side comparison of domestic BSE has been made with European BSE isolates. Between February 16-18, 2010, ARS researchers at Ames, Iowa, inoculated cattle with domestic and European BSE isolates. This study is expected to last at least 2 years before all animals will show signs of clinical disease and will provide the first direct comparison of these isolates and provide sufficient material for future studies of BSE. The start of research efforts on 24 milestones characterizing atypical versus classical BSE are dependent on tissues obtained at the completion of this animal study.

Production of cattle containing the rare 211K PRNP allele associated with genetic bovine spongiform encephalopathy (BSE). The 211K PRNP allele identified as being associated with genetic BSE had been identified in only 1 living animal, limiting the ability to actively study the impact of this allele on BSE. ARS researchers at Ames, Iowa, with cooperators at Iowa State University, produced 13 calves to date (about half containing this allele) using superovulation and embryo transfer. Production of these calves initiates a long-term animal study testing the hypothesis that this rare, naturally-occurring allele is a cause of genetic BSE in older cattle. This also enables ARS scientists to expand and preserve a unique scientific resource for the study of BSE as some of these calves are now being used to expand the pool of infectious BSE material from the Alabama 2006 BSE case, which is another resource in limited supply, and will ultimately allow proof of the existence of genetic BSE.

## 2. Other ARS Research activities also designed to do no harm:

Invasive species information portal: The National Agricultural Library National Invasive Species Information Center (invasivespeciesifno.gov) web site provides an information gateway to invasive species information; covering Federal, State, local and international sources.

Information management support to ITAP: The National Agricultural Library provides information management support for the Federal Interagency Committee for Invasive Terrestrial Animals and Pathogens (ITAP), a Federal scientific and technical interagency advisory group.

Overseas laboratories/quarantine facilities: Classical biological control is the use of natural enemies derived from a pest's point of origin. It offers the possibility for permanent, cost effective suppression of weeds and insect pests. The ARS Overseas Biological Control Laboratories (OBCL) are located in Australia, China, Argentina, and France and work as a cohesive network. Their collective mission is to identify, develop and ship natural enemies to stateside collaborators for use in U.S. programs designed to combat invasive species. Accordingly, they represent the beginning of a

pipeline of effective biological control agents and numerous stateside programs rely upon them. The ARS OBCL has a rich history of success in this regard, having contributed numerous biological control agents now in use across the U.S. ARS OBCL maintains formal collaborations with APHIS, the U.S. Forest Service, the U.S. Fish and Wildlife Service, the Bureau of Land Management, the Bureau of Indian Affairs, and many State Departments of Agriculture.

Related to this overseas work, ARS maintains quarantine facilities for insects and pathogens that meet Federal safety specifications to preclude pest introduction into the U.S. When beneficial insects arrive from overseas, they are carefully sorted, screened for parasites and reared or cultured within the quarantine facilities. ARS operates laboratories with quarantine facilities in Albany, California, Florida, Maryland, Mississippi, and Montana. Each quarantine facility uses a variety of traps, doors, entryways and sanitizing procedures to keep the pests secure until they are proven safe for release into the U.S.

### 3. Activities that are doing harm, and future agency actions to change them so that they do not continue to do harm.

None. As the principal in-house research agency for the United States Department of Agriculture, ARS conducts research to develop and transfer solutions to agricultural problems of high national priority. ARS scientific studies provide data and develop tools that enable America to change potentially harmful actions into those that do no harm while still meeting the challenge posed by invasive species.

## B. National Institute of Food and Agriculture (NIFA) (previously named the Cooperative State Research, Education and Extension Service (CSREES)

### 1. Activities to do no harm

a. <u>Technical Advisory Group for the Biological Control of Weeds:</u> NIFA is a member of the Technical Advisory Group (TAG) for the Biological Control of Weeds. This advisory group is made up of representatives from various Federal

agencies that evaluate candidate biological control agents for their economic, environmental, and ecological safety. Should the candidate biocontrol agents receive approval for release against a given target weed, this helps ensure that harmful non-target effects from the natural enemies are minimized. TAG advises APHIS.

b. National Animal and Plant Diagnostic Laboratory Networks: The safety of U.S. plant and animal production systems depends on our ability to rapidly identify foreign pathogens and other pests, whether introduced intentionally (through bio-terrorism) or unintentionally. NIFA has established two national networks of existing diagnostic laboratories to rapidly and accurately detect and report pathogens of national interest and to provide timely information and training to state university diagnostic laboratories.

The National Plant Diagnostic Network is led by five regional laboratories (Cornell University, University of Florida, Michigan State University, Kansas State University, and University of California-Davis) and one support laboratory (at Texas Tech. University).

The National Animal Health Laboratory Network (NAHLN) is led by 12 Core Laboratories and 58 total laboratories (receiving training/reagent/exercise support and being linked) in 43 states. NIFA is currently helping labs (other than the 12 core laboratories) with funding to set up electronic (secure, standards-based) messaging regarding FAD findings. These facilities will help to link growers, field consultants and other university diagnostic labs to coordinate regional detection and provide inter-regional communication in the event of an outbreak. For more information on the NAHLN see http://www.aphis.usda.gov/animal_health/nahln/downloads/NAHLNBriefingCurrent.pdf

## 2. Other Agency Activities, also designed to do no harm

a. Integrated Pest Management**:** Section 15 of the Federal Noxious Weed Act of 1974, and the Executive Order 13112 on Invasive Species (signed in 1999) direct Federal agencies to use an integrated pest management (IPM) approach for the management of undesirable plants on Federal lands using all available tools, including: education; preventive measures; cultural, mechanical, physical, biological and chemical control; and general land management practices such as revegetation, manipulation of livestock or wildlife grazing, and improvement of livestock and wildlife habitat.

Integrated Pest Management provides a sustainable approach to managing pests by combining biological, cultural, physical and chemical tools in a way that minimizes economic, health and environmental risks. The adoption and utilization of IPM is being encouraged through other legislative authorities within Federal departments. For example, US Code (Title 7, Chapter 6, Subchapter II, Sec. 136r-1. Integrated Pest Management**)** states: "The Secretary of Agriculture, in cooperation with the Administrator, shall implement research, demonstration and education programs to support adoption of Integrated Pest Management." It further states "Federal agencies shall use Integrated Pest Management Techniques in carrying out pest management activities and shall promote Integrated Pest Management through procurement and regulatory policies and other activities. IPM is also being encouraged across Federal agencies within the Department of the Interior.

Because of the complexity of economic, social, and environmental issues associated with invasive species management, and the biological and ecological attributes associated with each particular invasive species, programs that are based on a combination of technologies tend to be most successful and sustainable. As indicated in the National Invasive Species Council's (NISC) National Invasive Species Management Plan of 2001, the IPM

approach considers the best available scientific information, updated target population monitoring data, and the environmental effects of control methods in selecting a range of complementary technologies and methods to implement to achieve a desired objective. Some of the factors to consider in selecting control methodologies include environmental compatibility, efficacy, cost-effectiveness, inter-compatibility of different types of control measures, practicality and safety. The adoption of an IPM approach for invasive species management will certainly help minimize harm to the environment, human health and wildlife.

**3. Activities that are doing harm and future agency actions to change them so that they do not continue to do harm**

a. Pesticide use that has negative impacts: Conventional pest management strategies using pesticides are still emphasized in the management of invasive species with potential negative side effects to humans, the environment and wildlife. NIFA is helping to facilitate the adoption of an Integrated Pest Management Roadmap (IPM Roadmap) that will certainly help minimize harm to non-target species and the environment.

The goal of the IPM Road Map is to increase nationwide communication and efficiency through information exchanges among Federal and non-Federal IPM practitioners and service providers including land managers, growers, structural pest managers, and public and wildlife health officials. Development of the Road Map for the National Integrated Pest Management (IPM) Program began in February 2002, with continuous input from numerous IPM experts, practitioners, and stakeholders. The Road Map identifies strategic directions for IPM research, implementation, and measurement for pests in all major settings, throughout the nation. This includes pest management for areas including agricultural, structural, ornamental, turf, museums, public and wildlife health pests, and encompasses terrestrial and aquatic invasive species.

The goal of the National IPM Program is to increase the economic benefits of adopting IPM practices and to reduce potential risks to human health and the environment caused by the pests themselves or by the use of inappropriate pest management practices.

b. Pest Management Grant Programs: NIFA has several competitive grant programs designed to emphasize IPM, while reducing pesticide residues on food and in the environment. These include the Risk Avoidance and Mitigation Program, Crops at Risk Program, Pest Management Alternatives Program, Integrated Organic Program, Methyl Bromide Transitions Program, Regional IPM Competitive Grants Program, and the IPM Centers. The emphasis of IPM and bio-based pest management in these NIFA competitive grant programs will certainly help minimize harmful side effects to non-target species and the environment when these strategies are used in invasive species management.

c. $IPM^3$ Training Consortium for Federal Employees: NIFA, in collaboration with Land Grant Universities and other Federal Agencies, has facilitated the development of an IPM distance education platform to provide IPM training to Federal workers involved in pest management issues and activities. Increasing the quality and consistency of IPM training among Federal agencies and their adoption of an IPM approach for invasive species management will certainly help minimize harm to the environment, to human health, and to wildlife. For more information on $IPM^3$ please visit the following website: www.umn.edu/ipm3.

d. Pest Information Platform for Extension and Education (PIPE): PIPE is a reporting and tracking system, developed collaboratively with the USDA Risk Management Agency, to manage pest and disease information flow via the Web. The PIPE system provides real-time useful information to U.S. crop producers, and a “one stop shopping” center for timely, unbiased, national, and local pest information. PIPE

fosters good farming practices by encouraging growers to: avoid unnecessary or ill-timed chemical applications; use the proper control tactics with the proper timing to manage crop loss risk; and document practices for crop insurance purposes. The PIPE system for soybean rust saved growers hundreds of millions of dollars in 2007 by providing real-time information that enabled the growers to avoid unnecessary chemical applications. Additional active ipmPIPE components include: soybean aphid, legume diseases, curcurbit downy mildew, pecan, and southern corn rust.

## C. Economic Research Service (ERS)

### 1. Activities to do no harm

ERS is the main source of economic information and research from the U.S. Department of Agriculture. ERS research informs and enhances public and private decision-making on economic and policy issues related to agriculture, food, natural resources, and rural development.

a. Program of Research on the Economics of Invasive Species Management (PREISM): ERS initiated a new program of work in fiscal year 2003, the Program of Research on the Economics of Invasive Species Management (PREISM), to examine the economic issues related to managing invasive species in increasingly global agricultural markets. Through PREISM, ERS primarily funded extramural research through a competitive awards program that focuses on national decision making concerning invasive species of agricultural significance or affecting, or affected by, USDA programs. In addition to ERS-led analyses of invasive species issues, ERS has disbursed $6.8 million through the competitive awards program to 45 recipients, including universities, other USDA agencies, and private non-profit institutions, for research on the economics of invasive species during FY 2003 to FY 2008. About $1.1 million per year were allocated for extramural agreements in FY 2005 and FY 2006, while $950,000 was allocated in FY 2007 and $970,000 in FY 2008. No Funds were

allocated in FY 2009 and FY 2010. ERS also organized annual workshops from 2003 to 2009 to provide a forum for dialogue on economic issues associated with agricultural invasive species.

Following are some preliminary findings from PREISM-funded research projects:

- Prevention and management resources should be allocated to species and strategies with the highest return (in terms of damage reduction over time). Ideally, marginal benefits and costs should be equal across species and strategies.

- Decision-support tools that follow sound economic principles and reveal underlying scientific assumptions and value judgments provide a basis for expert and stakeholder involvement in decision-making and promote efficient allocations of funds.

- Optimal invasive species management strategies depend upon the stage of the invasion and associated rates of growth and spread. Eradication may be optimal for small invasions; reduction to a containment level for larger invasions. If eradication is feasible, the effort will reduce discounted damages more if it occurs early when populations are small. Delays result in more damages. If total cost increases rapidly as population increases, eradication when the population is small followed by prevention may be the best strategy.

- Under-funded eradication or management efforts can be cost-ineffective or wasteful, with little or no effect on invasive species growth and total damage. Higher initial expenditures can reduce long term damages and control costs, even if the species is not eradicated.

- For established invasive species infestations, per unit costs of removal can increase as populations decrease or become more isolated, making complete eradication difficult or cost-

inefficient. In some cases, accommodation to low levels of invasion is economically preferable to the high cost of eradication. The higher is the cost of removal, the larger the population that will be accommodated.

- Higher invasive species infestation or population growth rates reduce benefit-cost ratios of control efforts, and at high enough rates, control might not be worthwhile. If population has surpassed that of maximum growth rate, the best strategy could be a pulse-like effort that drives populations below a critical population level and growth rate, followed by containment strategy.

- Probability of occurrence maps for invasive weeds based on GIS and other inventory or survey data and related population growth rates can improve weed management efficiency by reducing: 1) costs by targeting sites to monitor invasiveness, and/or 2) damage by initiating control of highly invasive populations before they spread.

- Coordination of regulations across U.S.-Canada, State, and provincial boundaries could: 1) more effectively reduce the cross-border spread of exotic horticultural plants that become invasive, and 2) reduce incentives for cross-border firm relocations to take advantage of more lenient regulations.

- Ecological and agronomic differences influence cross-State differences in noxious weed and weed-seed lists, but stakeholder lobbying also has significant effects.

**2. Other Agency Activities, also designed to do no harm**

ERS is not engaged in any activities that do harm.

**3. Activities that are doing harm and future agency actions to change them so that they do not continue to do harm**

None.

## II. USDA Regulatory and Resource Management Agencies

### A. Animal and Plant Health Inspection Service (APHIS)

#### 1. Activities to do no harm

"Protecting American agriculture" is the basic charge of the U.S. Department of Agriculture's (USDA) Animal and Plant Health Inspection Service (APHIS). APHIS provides leadership in ensuring the health and care of animals and plants. The agency improves agricultural productivity and competitiveness and contributes to the national economy and the public health. APHIS has major regulatory authority to implement action programs to achieve these responsibilities. For more detailed information and up to date highlights of program activity, please visit the APHIS Web Site (http://www/aphis.usda.gov/).

a. Invasive Species Prevention Programs: Specifically the APHIS mission, stated in its current strategic plan, is to protect the health and value of American agriculture and natural resources. To carry out this mission, APHIS works to achieve two interdependent goals:

- Safeguard the health of animals, plants, and ecosystems in the United States (U.S.)
- Facilitate safe agricultural trade

It does so through a system of interdependent objectives addressing exclusion (i.e., prevention), detection, emergency response, management, trade issue resolution, and capacity building. These areas correspond closely to elements of the 2001 National Invasive Species Management Plan.

APHIS tries to ensure that other entities in the private and public sectors, including other Federal agencies, "do no harm" by introducing or spreading invasive species. APHIS prevention programs – a comprehensive set of risk-based regulations and enforcement efforts -- are directed at animals, plants, and their products that may bring invasive species or be pathways for the introduction of invasive species. As such, the

Agency addresses both unintentional and intentional introductions of invasives. A description of some of the applicable regulations follows.

1. Regulation of certain animals and animal products: APHIS regulates, as set forth in 9 CFR parts 91 through 99, the importation of animals and animal products to guard against the introduction of animal diseases into the U.S. in accordance with the Animal Health Protection Act.

2. Regulation of certain plants and plant products: Regulations contained in 7 CFR part 319 prohibit or restrict the importation of plants, plant parts, and plant products into the U.S. in accordance with the Plant Protection Act. APHIS enforces the part 319 regulations and considers requests to amend the part 319 regulations to allow the importation of plants, plant parts, or plant products that are not currently allowed importation under the regulations. The requirements apply to many commodities, including nursery stock.

3. Listing of noxious weeds:
Under the authority of the Plant Protection Act, APHIS regulates, in 7 CFR parts 360 and 361, the importation and interstate movement of plants and plant products that may be noxious weeds, i.e., plants that can directly or indirectly injure or cause damage to crops, livestock, poultry, or other interests of agriculture, irrigation, navigation, the natural resources, public health, or the environment.

b. Plant Epidemiology and Risk Analysis Laboratory (PERAL): PERAL is a diverse group of scientists and professionals comprising the primary office in Plant Protection and Quarantine (PPQ) for pest risk analysis. PERAL is responsible for providing essential scientific support to risk-based policy making across a broad range of phytosanitary issues. The staff uses scientific principles, procedures and evidence to analyze issues relevant to safeguarding plant health from the threats of

harmful exotic pests of cultivated and natural plant systems. This includes most risk analyses required by PPQ for pests, Commodities, and pathways but it does not currently include risk analyses associated with plant pest permits, genetically modified organisms, or Federal Noxious Weeds.

PERAL serves a wide range of functions within PPQ. The overarching responsibility is to provide comprehensive, accurate information in support of the decision making process ensuring that resulting actions are the most appropriate and "Do No Harm". For more in-depth information regarding PERAL, please visit http://cphst.aphis.usda.gov/plant health/cphst/peral.shtml

A good example of one of these functions is the New Pest Advisory Committee: The New Pest Advisory Group (NPAG) is located in the APHIS Center for Plant Health Science and Technology (CPHST), Plant Epidemiology and Risk Analysis Laboratory (PERAL). The overall goal of NPAG is to safeguard American agriculture and natural resources. The NPAG assesses new and imminent exotic plant pest introductions into the U.S. to recommend appropriate Plant Protection and Quarantine's (PPQ) policy and actions to respond to the potential threat posed by such pests. In this case a pest is defined as: *Any species, strain or biotype of plant, animal or pathogenic agent injurious to plants or plant products* [FAO, 1990; revised FAO. 1995; IPPC, 1997].

NPAG may address pests in many taxa including arthropods, plant pathogens, mollusks and weeds. It determines whether the pest is a present or an imminent threat, and if the pest meets the definition of a quarantine pest. If the pest meets the definition, NPAG may convene an ad hoc panel of Subject Matter Experts from PPQ, other Federal, state, and university sources with regulatory and scientific expertise for that particular exotic pest. Through literature searches, data sheet preparation and discussion with the panel, NPAG provides findings and recommendations via the NPAG Report to the APHIS Deputy Administrator and the APHIS Executive Team

(represented by PPQ's management) in response to the pest introduction.

c. APHIS Wildlife Services (WS) Activities

Nonnative, invasive species can be devastating to ecosystems where a lack of natural enemies and competition for resources can allow these species to thrive, wiping out other native wildlife in the process. APHIS WS' efforts target these introduced and invasive species. Invasive species of concern include brown tree snakes (BTS), Gambian rats, nutria, Coqui frogs, pigeons and starlings, house sparrows, feral swine and Burmese pythons.

1. Feral swine are an introduced species that pose a number of threats to humans, livestock and wildlife. Among these threats is the ability of feral swine to harbor a variety of federally regulated pathogens whose presence would result in severe economic loss to livestock industries. Estimates of economic losses from feral swine to agriculture and the environment average $800 million annually. Feral swine have established populations in 38 states and are spreading rapidly. WS removed 28,472 swine in 29 states in FY 2008.

2. European starlings are an invasive species that invade livestock facilities, eating and defecating in feed bins. This fouling causes severe economic losses to the farmer and transmission of disease and loss of production in the animals. Estimates of economic losses due to starlings range from $800,000 - $4,137,119 annually in the U.S. WS removed 1,703,697 starlings from livestock facilities in 42 states in FY 2008.

3. Brown tree snakes have eliminated 10 of the 13 native bird, most lizard, and bat species on the island of Guam, are responsible for large economic losses from damaged electrical lines and resultant power outages, and pose a hazard to human safety from bites. APHIS continued to prevent the unintentional introduction of the BTS from Guam to other Pacific Islands, Hawaii, and the continental

United States in FY 2008. The Agency intercepted 12,212 BTS on Guam or near ports of exit. APHIS WS National Wildlife Research Center scientists at the Fort Collins, Colorado headquarters, conducted an economic assessment of a hypothetical translocation of the BTS from the Territory of Guam to the Hawaiian Islands. The total annual projected economic impact of the translocation of BTS to Hawaii was estimated to fall within the range of $473 million to $1.8 billion. These projections underscore the value of a BTS interdiction and control program on Guam.

4. The Gambian rat is a very large rodent native to northern Africa. Gambian rats can harm livestock species and habitats, damage agricultural crops, consume livestock feed, and are associated with a variety of pathogenic diseases that could be spread to humans, livestock, and wildlife. APHIS continues to work with the Florida Fish and Wildlife Conservation Commission, U.S. Fish and Wildlife Service, South Florida Water Management District, and the Florida Park Service to move toward the eradication of the Gambian rat from the Florida Keys. Removal methods have been successful and rat numbers are down significantly over previous years.

5. Nutria are large, semi-aquatic rodents native to South America, but are now established in 17 states and cause extensive damage to wetlands, agricultural crops, and structural foundations such as dykes and roads. The rodents may also threaten human health and safety and serve as a reservoir for tularemia and other diseases. APHIS is leading the first large-scale North American effort to eradicate a mainland population on the Delmarva Peninsula in Maryland where the rodents have devastated coastal Chesapeake Bay marshes. In cooperation with the Department of Interior's Fish and Wildlife Service, Maryland Department of Natural Resources, U.S. Geological Survey (USGS), Tudor Farms (a 6000-acre private wildlife management area) and 300 private landowners, APHIS has completed the

initial nutria removal from more than 150,000 acres of coastal marsh in Maryland. The Agency is now expanding the search for established populations in major tributaries leading into the region. The Agency's wildlife specialists have developed and refined new removal techniques and have partnered with USGS to develop new detection and monitoring techniques including remote triggered cameras, call-back surveys, and other means of detecting low density populations. Through careful population monitoring, APHIS has successfully prevented the re-infestation of this area, and marsh grasses and native muskrat populations are quickly recovering throughout the previously-impacted area.

In addition to the species highlighted, APHIS provides assistance to the general public upon request to resolve damage caused by invasive species. Last Fiscal Year, APHIS provided direct control assistance to resolve damage caused by 14 of the 23 bird, mammal, and reptile species identified by the World Conservation Union (IUCN) as being among the top 100 invasive species in the world. These species included: BTS, giant toad, Coqui frog, red-vented bulbul, common myna, European starling, nutria, house mouse, roof rat, small Asian mongoose, feral swine, cats and goats.

d. APHIS Veterinary Services (VS) Activities

1. National Animal Health Laboratory Network (NAHLN) is a state-federal cooperative effort including the APHIS National Veterinary Services Laboratories, which provide reference and confirmatory laboratory services including training, proficiency testing, and prototypes for diagnostic tests. The State/University laboratories in the NAHLN perform routine diagnostic tests for endemic animal disease as well as targeted surveillance and response testing for foreign animal diseases. The network will assist in early detection and rapid, scalable response to an exotic animal disease.

2. Cattle fever is a severe and often fatal disease of cattle transmitted by two species of tick. The ticks were

eradicated from the continental US in 1943, with the exception of a buffer zone between Texas and Mexico. An increase in movement of deer and stray livestock across the border has led to increased fever tick infestations in recent years despite a partial tick control border fence, livestock movement quarantines, and tick treatments for cattle and deer. In response, APHIS proposes extending and filling in gaps in existing fencing in three Texas counties. APHIS will prepare an EIS to examine the potential environmental effects of the proposed action and alternatives to that action.

3. The APHIS response to the invasion of pandemic influenza included investigation and characterization of novel swine influenza isolates, and preparing and distributing a pre-qualified 'master seed virus' of pH1N1 to veterinary biologics manufacturers, greatly reducing the time to make the vaccine available to US swine producers.

4. APHIS partnered with the State and Federal agencies in Florida to record information on exotic pet sales outside of traditional pet stores, and talk with owners about animal health. A public meeting was also held. As a result, APHIS located 17 venues where exotic pets are marketed, 13 of which were unknown to animal health officials, and most of which are in close proximity to traditional livestock operations.

5. Foot and Mouth Disease is the most communicable disease known, and is exotic to the US. APHIS activities have recently included vaccine and pen-side diagnostics studies, and characterizing the pathogenesis and clinical signs in feral swine.

6. Rift Valley Fever is an arthropod-borne zoonotic disease (infects humans and non-humans). APHIS activities have recently included diagnostic test validation and geospatial collaborations.

7. APHIS is partnering with university and industry entities to increase the value of its disease spread modeling programs by adding livestock movement data. An exotic or emerging animal disease would likely move most quickly through current production-oriented animal movement.

### 2. Other Agency Activities, also designed to do no harm

Program protocols: APHIS also follows protocols to ensure that its own activities and those of its State cooperators, carried out to exclude, detect, diagnose, control, and eradicate invasive species, do not contribute to the problem. These ongoing efforts include, in a general sense, agency personnel adherence to established biosafety procedures in programs to detect, diagnose, and conduct control operations for plant and animal diseases and pests, both in laboratories and in the field; and assessment, in advance, of the probable impact of the use of biocontrol agents in programs to control invasive species.

### 3. Activities that are doing harm, and future agency actions to change them so that they do not continue to do harm

None. APHIS actions are consistent with the "DO NO HARM" objective of the Presidential Executive Order on Invasive Species.

## B. Natural Resources Conservation Service (NRCS)

### 1. Activities to do no harm

The NRCS is well aware of the past, the present, and the potential future harm to the private lands in the U.S. from invasive species. The negative environmental and economic impacts of invasive species continue to be a large and growing problem for our Nation's private landowners.

The primary invasive species focus for NRCS has been on terrestrial and aquatic invasive plants. Invasive plants have had large negative environmental impacts upon the intended uses of many privately owned lands and wetlands in the U.S. There have also been large negative economic impacts associated with the costs of invasive plant control. Invasive plants compete for soil nutrients and water in croplands and wild lands and often require the use of herbicides, biological control agents, or innovative control techniques. Invasive plants, often of poor forage quality, may out-compete native plants in grazing lands and wild lands rendering large acreages no longer useful for supporting livestock or wildlife. Invasive aquatic plants rapidly spread in water bodies and wetlands, removing the open water component necessary for many wildlife species. Of particular concern are the negative impacts from invasive plants, invasive invertebrates, and pathogens upon populations of native and introduced pollinators and their habitats as well as upon native threatened or endangered species and their habitats. The invasive species could have devastating effects on desirable cropland and wild land plants and animals.

a. Publication and Revision of Agency Invasive Species Policy: NRCS published its NRCS Invasive Species Policy in November 2004 (available at **and revised it in July 2010. This policy is available at http://policy.nrcs.usda.gov/scripts/lpsiis.dll/GM/GM_190_414.htm.** The policy addresses the invasive species responsibilities at all levels (e.g., National Headquarters, Regional, State, and Field offices) of the agency. It requires awareness by NRCS employees of the presence of invasive species and potential problems associated with them. It requires NRCS to work with partners and to use its human and financial resources for control, suppression, and/or eradication of invasive plants. The policy also requires that native plant species be used in vegetative conservation practices unless it can be demonstrated that no native species can achieve the desired conservation goals, or the desired native species is not available in the quantity required. Interim use of non-native

species is allowed to provide the conservation function desired until native species can be established.

b. Assisting in the control and eradication of invasive plants: NRCS provides U.S. private landowners with financial and technical assistance to control and/or eradicate invasive plants in an effort to maintain the desired vegetation (e.g., food crops and forage), to maintain the desired characteristics of the land (e.g., wetland open water), and to diminish invasive plants spreading to neighboring lands. NRCS frequently partners with local and regional weed control organizations for control of weeds on and off private lands. The agency encourages the use of integrated pest management (IPM) which may involve appropriate herbicides when necessary, the use of approved biological control organisms, and innovative cultural control methods for specific problems (e.g., black plastic). NRCS has placed increased emphasis upon the protection of wildland habitats for pollinators and other wildlife

Landowners that participate in some of the easement programs of NRCS (e.g., Conservation Reserve Program (CRP), Wetlands Reserve Program (WRP)) are required to control invasive plants that might infest the easement lands. CRP and WRP participants may receive some financial assistance to maintain these lands free of invasive plants. The Wildlife Habitat Incentives Program, Environmental Quality Incentives Program and the Conservation Stewardship Program also provide technical and financial assistance to help private landowners control invasive plants.

c. NRCS Conservation Practice Standards: NRCS has created a toolbox of 170 practice standards that provide guidance for applying conservation technology on the land and that set the minimum levels for acceptable application of the technology. These practice standards undergo periodic review for incorporation of new technology (generally every 5 years). Emphasis continues to be placed upon the identification and consideration of the invasive qualities of recommended vegetation, the use of native vegetation, and the protection and enhancement of pollinator habitat.

d. The NRCS Plant Materials Centers (PMCs): The 27 PMCs nationwide cultivate and provide seed stock of plants that are used for vegetative conservation practices within the geographical region served by each PMC. The PMCs encourage use of native plants, particularly source-identified plants, for restoration, reclamation, and conservation practice uses. The Plant Materials program uses an Environmental Evaluation to assess the potential invasiveness of plants being considered for release. If the potential for invasiveness is too great, other plants considered less invasive for the particular environmental conditions are recommended.

The PMCs also used the Environmental Evaluation to review all prior NRCS conservation plant releases. For plant releases that were determined to be invasive or otherwise environmentally harmful, the PMCs discontinued their production. Once a PMC discontinues a plant release, the NRCS plant materials specialists work with the appropriate states to remove the invasive plant releases from NRCS State standards and recommendations so that plant is not recommended in the future.

**2. Other Agency Activities, also designed to do no harm**

a. PLANTS Database: The information about plant materials available through the PLANTS database (http://plants.usda.gov) is useful to conservation professionals and the public in determining beneficial plants that do well within a particular geographical location. It also has information on plants which should not be planted within a particular environment (e.g., Federal and State noxious weed lists). The database information provides help to assess the potential invasiveness of specific plants. The PLANTS database has over 650 fact sheets on-line line and provides services through over 70,000 user sessions per day. It encourages the use of native plants in conservation practices. Future capabilities will include information about the pollinators upon which specific plants are dependent, and recommended forage to encourage specific pollinators.

**3. Activities that are doing/have done harm, and agency actions to change them so that they do not continue to do harm**

a. Recommending invasive plants in conservation plans. During the "Dust Bowl" days of our nation, immediate action was necessary to mitigate excessive wind and water erosion of our nation's soils. Unfortunately, one of the mitigation tools that worked effectively was the use of non-native plant materials, some of which became invasive and presently are among the invasive plant materials we are trying to control. The use of the Environmental Evaluation by the PMCs before recommending specific plant materials for conservation is proving to be beneficial to avoid present and future problems of this kind. Also, encouraging the use of locally-acquired native plants whenever they can meet the conservation needs is enhancing awareness to NRCS state and field offices about invasive species problems and NRCS responsibilities.

The implementation of the NRCS Invasive Species Policy has made clear to all levels of the agency the responsibilities to respond to invasive species problems, and to minimize or avoid future invasive species problems.

The state-specific Field Office Technical Guides are technical guidance information for the specifics of each conservation practice standard within the specific State. Technical Guides may, in some cases, still recommend the use of plant materials that may become invasive. NRCS has conducted and continues a review of all vegetative conservation practice standards to identify where this situation exists, and to work with the appropriate PMCs and State Plant Materials Specialists to recommend other appropriate and non-invasive plant material.

b. Use of herbicides or other methods that may have detrimental effects on native pollinators: The treatments recommended in some conservation practice standards for invasive plants may, in some cases, include the use of

herbicides or other methods that may have detrimental effects directly or indirectly (e.g., habitat destruction) on native pollinators. NRCS continues to review and to revise all practice standards to identify such methods, and to recommend revisions that minimize or eliminate negative impacts to native pollinators. NRCS is developing a module within the PLANTS database that identifies specific plant-pollinator relationships and encourages the use of "pollinator friendly" plants in agricultural and wild land situations.

## C. U.S. Forest Service (FS)

### 1. Activities to do no harm

a. Invasive activities on 193 million acres of National Forests and Grasslands: The USDA Forest Service (FS) is increased activities to prevent, control, and eradicate aquatic and terrestrial invasive species (including plants, pathogens, vertebrates, and invertebrates) across the National Forest System lands, 193 million acres. In FY 2010 the National Forest System (NFS) treated nearly 420,000 acres of lands and waters infested with invasive species, of which approximately 110,000 acres targeted non-plant invasives. In 2010, the national average outcome performance level for the percentage of priority acres restored/protected on NFS was approximately 78% treatment efficacy.

b. Supporting establishment of Cooperative Weed Management Areas (CWMA): The National Forest System programs increased support for a national initiative with Federal, state, and local partners to expand the establishment of CWMA across all states. Using models from areas of the country where they have been effective, the USFS continued to support a CWMA development and mentoring program.

c. Policy on invasive species management in national forests: The development of a new Forest Service Manual (policy) on invasive species management for the National Forest System

continued throughout 2010, with the expectation to release the proposed directive in the Federal Register for public comment by December 31, 2010. Plans continue on the development of an accompanying NFS Invasive Species Management Handbook to support the policy guidance.

d. Training, funding and technology for invasives work: NFS conducted and/or provided technical and financial support for numerous invasive species training workshops, educational programs, community outreach activities, and developing technology for invasive species management solutions.

e. Invasives Species Management Record Keeping and Reporting. The National Forest System invasive species data management applications were redesigned and improved to include key aspects of invasive species treatment and inventory work, as well as new program performance measures. Guidance and 2010 program direction to Forest Service regions, national forests, and forest districts was provided through several channels and available on-line. Improvements include the use of personal data recorders for quicker collection of field data (spatial and tabular) on all taxa of invasive species. Forest Service data applications continue to evolve as technology and security requirements change.

f. FS Invasive Species Issue Team: The FS has an active Washington Office Invasive Species Issue Team. It has representatives from four program areas in the FS: National Forest System; Research & Development; State & Private Forestry (SPF); and International Programs. The team discusses invasive species issues and problems and addresses them in a comprehensive and collaborative way among agency programs.

g. USFS web site on invasives: USFS established a national website on invasive species. It provides user information on FS activities related to invasive species, policy, authorities, news and emerging issues. The site provides key contact information for invasive species program managers, access to cooperative projects and research, geographic information, species profiles,

and techniques for preventing and controlling a wide variety of species. The website is http://www.fs.fed.us/invasivespecies/

h. FS activities in support of NISAW: The National Forest System provided continuing support for the annual National Invasive Species Awareness Week (NISAW) activities in Washington, DC. Many Forest Service local and regional offices conducted weed education and awareness activities with partners at the local level at the same time as the NISAW activities took place in DC.

i. Completion of OIG Audit on the Invasive Species Program: On September 30, 2010, Assistant Inspector General for Audit, Gil H. Harden formally notified the Chief of the Forest Service that the USDA Office of Inspector General (OIG) had reached management decision on all recommendations in the subject audit report (Audit Number 08601-7-AT). The "Achievement of Management Decision" document articulated eleven (11) final recommendations for improvements in the Forest Service invasive species program, inclusive of each of the key Deputy Areas (SPF, NFS, and R&D) involved in the program. For each OIG recommendation, the Forest Service provided it's response and corresponding timetable for completion of the final action(s). OIG recommendations included a number of major program changes needed to increase accountability and transparency in the program; including developing and implementing controls for reporting accurately how much the agency is spending to combat invasive species both locally and nationally, and implementing comprehensive national policy on invasive species management through the Forest Service Manual. The Forest Service will begin implementing actions agreed upon in the Achievement of Management Decision document to address the issues and recommendations identified by OIG. The first action to be implemented, the release of the draft Forest Service Manual 2900 (Invasive Species Management) in the Federal Register for public comment, will occur by December 31, 2010.

j. Research on invasive species

Several USFS employees have been participating in a “think tank” funded by The Nature Conservancy at UCSB’s National Center for Environmental Analysis and Synthesis.

The group’s first paper (Aukema, et al., Bioscience 60:886-897) describes the historical accumulation of non-native forest insect pests and diseases. This paper is the first step in an economic impact analysis to inform regulatory and policy decisions. More than 450 non indigenous insects and at least 16 pathogens have colonized forest and urban trees since European settlement. Approximately 2.5 established non indigenous forest insects per year were detected in the United States between 1860 and 2006. At least 14% of these insects and all 16 pathogens have caused notable damage to trees. Whereas most guilds’ introduction rates were fairly constant, detections of insects that feed on phloem or wood have increased markedly since 1980. The increase in wood borers is likely due to the rise in containerized freight in the last few decades. Container shippers often use wooden pallets, crates and dunnage to brace equipment, marble and other large commodities. This wood packaging material is often made of low quality wood that can contain forest pests.

The group also published “Cost of potential emerald ash borer damage in U.S. communities, 2009-2019”, which shows that local governments and homeowners actually will bear most of the costs (predicted at $1 billion USD/yr over the next 10 yrs) for this invasive insect. Ecological Economics 69: 569-578.

Published two volumes on “Advances in Threat Assessments and their Application to Forest and Rangeland Management” (GTR-PNW-802) on disturbances of all types, including invasive pests at the landscape level. Relevant papers include: Representing Human-Mediated Pathways in Forest Pest Risk Mapping; Decision making under risk in invasive species management: risk management theory and applications; Review of Methods for Developing Regional Probabilistic Risk Assessments, Part 2: Modeling Invasive Plant, Insect, and Pathogen Species.

Provided the science review to USDA-APHIS on proposed international standards for plants for planting and criteria for adding new treatments to the wood packing standard.

Armillaria root pathogens have a worldwide distribution and can infect most woody plant species. Although many geographic areas possess native Armillaria pathogens, movement of these pathogens to new geographic areas poses an invasive pathogen risk. National and international collaborations with RMRS researchers are providing DNA-based diagnostics to identify Armillaria species in Mexico (Cannon and others 2008; Kim and others 2010), Hawaii (Hanna and others 2007), and Alaska (Klopfenstein and others 2009b). These surveys will provide baseline information for managing other potentially invasive Armillaria pathogens. Bioclimatic modeling is then applied to identify which areas have a suitable climate for each Armillaria pathogen. New approaches are being developed to identify areas with a projected suitable climate for these pathogens under various climate change scenarios (Klopfenstein and others 2009a). Identifications will become more refined as the present distribution of Armillaria species becomes better documented.

Venette et al. published “Pest Risk Maps for Invasive Alien Species: A Roadmap for Improvement”. Bioscience 60: 349-362.

Publication on measurement and estimation of plant species that enables nationally consistent estimates of invasive plant abundance. Data is publicly available on the web at http://fia.fs.fed.us/tools-data/default.asp

USFS R&D is working to improve our use of internet sites to disseminate research results. Examples include:

NRS invasive species website contains information on key pests, (ie EAB, HWA, ALB, etc.) --- http://nrs.fs.fed.us/disturbance/invasive_species/

Southern Research Station Forest Threat accomplishments website ---
http://www.srs.fs.usda.gov/science/threats_accomp.html

Rocky Mountain Research Station website --
http://www.fs.fed.us/rm/wildlife/invasives/

Pacific Southwest Research Station websites --
(http://www.fs.fed.us/psw/topics/insect_disease/
*http://www.fs.fed.us/psw/topics/invasives/strawberryguava/ &*
http://www.fs.fed.us/psw/topics/invasives/strawberryguava/)

Pacific Northwest Research Station website --
(http://www.fs.fed.us/pnw/invasives/index.shtml)

**2. Other Agency Activities, also designed to do no harm**

a. Video series on Best Management Practices for Prevention: Throughout FY 2010 the National Forest System invasive species program worked closely with partners to develop the third video in a DVD series on invasive species prevention best management practices. This third DVD outdoor recreationists is titled "Playing Smart Against Invasive Species – How to Enjoy and Protect the Great Outdoors". The new DVD is planned to be completed and released in November 2010.

b. Evaluation of vehicle washing activities: The National Forest System continues to support the evaluation of vehicle washing activities/systems/protocols with public and private partners to evaluate the effectiveness of existing systems and mechanisms. Evaluations are based on a scientific approach to quantify effectiveness and determine treatment quality for various scenarios. Long term objectives of the project include building better protocols and contract specifications, and ultimately better effectiveness at preventing the spread of invasive species by equipment and vehicles.

c. FS Performance measures for invasives: The FS revised the performance and accountability system measures for all

invasive species program activities, agency-wide. New performance management systems are in place. Field data is being collected in corporate data management applications. Program performance is outcome-driven and will emphasize the effectiveness of treatments. All NFS invasive species program performance outputs and outcomes were incorporated into the Forest Service Performance Accountability System for upward reporting in FY 2010.

d. Prevention language in FS contracts: National Forest System stations utilize specific invasive species 'prevention' language to include in project contracts (such as timber sales, road management, facility construction, etc.) that specify requirements to minimize invasive species infestations and spread on national forests and grasslands. In FY 2010, the NFS program expanded the use of prevention language in management contracts for maintenance of utility rights-of-ways and other operations.

**3. Activities that are doing harm, and future agency actions to change them so that they do not continue to do harm**

None.