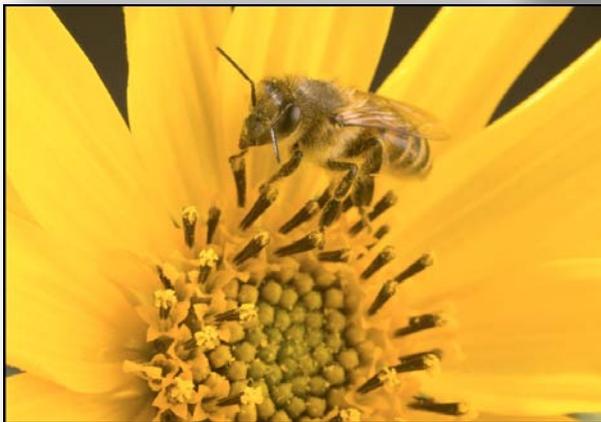


USDA
AGRICULTURAL RESEARCH SERVICE

NATIONAL PROGRAM 305
CROP PRODUCTION

ANNUAL REPORT FY 2010



National Program 305

CROP PRODUCTION

FY 2010 Annual Report

Introduction

National Program 305, Crop Production, supports research to develop knowledge, strategies, systems, and technologies that contribute to greater cropping efficiency, productivity, quality, marketability, and protection of annual, perennial, greenhouse, and nursery crops while increasing environmental quality and worker safety.

The Nation's rural economic vitality depends on the ability of growers to produce and market agricultural products – including food, fiber, flowers, industrial products, feed, and fuels – profitably, while at the same time enhancing the natural resource base upon which crop production depends. Future financial success depends on increasing productivity, accessing new markets for specialized products, developing technologies to provide new opportunities for U.S. farmers, and utilizing tools and information to mitigate risks and enable rapid adjustments to changing market conditions. The farm sector has great and varied needs driven by a wide variety of resource, climatic, economic, and social factors that requires an equally diverse array of solutions.

Contemporary cropping enterprises are complex and depend on highly integrated management components that address crop production and protection, resource management, mechanization, and automation. U.S. annual, perennial, and greenhouse (protected systems) crop production are all based on the successful integration of these components. The development of successful new production systems requires a focus on new and traditional crops; the availability and implementation of improved models and decision aids; cropping systems that are profitable and productive; production methods fostering conservation of natural resources; efficient and effective integrated control strategies for multiple pests; improved methods, principles, and systems for irrigation; improved mechanization; and reduced inputs – all while sustaining or increasing yield and quality.

Production systems must better address the needs of small, intermediate, and large farming enterprises including those using field-, greenhouse-, orchard-, and vineyard-based production platforms, through conventional, organic, or controlled environment strategies. Additionally, adaptation and/or development of technologies are required to ensure a sustainable and profitable environment for production agriculture. New technologies must address the need for lower cost, higher efficiency inputs that foster conservation of energy and natural resources, while maintaining profitability and promoting environmental sustainability.

In addition, declining bee populations and honey production require special attention. Over the past several years, a myriad of pests and potentially adverse cultural and pest management practices have been threatening many of the bee species required for pollination of a multitude of crops. Colony collapse disorder has now increased losses of the honey bee to a total mortality of over 30 percent. Also, as new crops or niches are introduced, there is an increasing need for non-honey bee pollinators for specific crops or protected environments.

NP 305 draws heavily on other ARS National Programs, universities, and industries in adapting and incorporating technologies, approaches, and strategies that enable the advancement of the Nation's agricultural industry and enhanced international competitiveness.

This National Program is divided into two main research components, with several sub-components:

- Component 1: Integrated Sustainable Crop Production
 - Subcomponent 1A: Annual Cropping Systems
 - Subcomponent 1B: Perennial Crops
 - Subcomponent 1C: Greenhouse, High Tunnel, and Nursery Production Systems
- Component 2: Bees and Pollination.
 - Subcomponent 2A: Honey Bees [*Apis*]
 - Subcomponent 2B: Non-*Apis* Bees

Below are National Program 305 accomplishments from fiscal year 2010, grouped by research component. This report is not intended to be a progress report describing all ongoing research, but rather a listing of accomplishments for the fiscal year, some of which are based on multiple years of research. As a result, not all research projects will reach an “accomplishment” endpoint each year.

ARS welcomes your input regarding our ongoing research programs. If you have any questions as you read the report below, please do not hesitate to contact either of the co-leaders for NP 305, Kevin Hackett (Kevin.Hackett@ars.usda.gov) or Sally Schneider (Sally.Schneider@ars.usda.gov).

Component 1: Integrated Sustainable Crop Production

ANNUAL CROPS:

Plants developed from organically produced bell pepper transplants do well under organic and conventional production. The method used to produce bell pepper transplants could affect subsequent plant development. ARS scientists in Lane, Oklahoma, evaluated how plants responded when transferred to the field from a growing medium inoculated with beneficial microbes during transplant production. The researchers inoculated a growing medium in the greenhouse with beneficial microbes and produced transplants using both organic and conventional methods. Seedlings were transferred to the field for further evaluation in two soil types, with planting occurring at various planting dates, and with use of organic and conventional production methods. The findings show that yield was affected by planting date (late summer planting was better than spring planting) and soil type (sandy soil was better than loamy soil). Inoculation with microbes during transplant production did not affect yield. Yield from organically produced plants was equivalent to that from conventionally produced plants. Nutrient content of fruit was not affected by inoculation, was similar between organic and conventional production methods, but was lower on plants grown on loamy than those grown on sandy soil. This information is useful to those using organic production methods for bell peppers and to those wishing to reduce synthetic inputs in a conventional production system.

Hand-held machine for assessing two-spotted spider mite damage in cotton. The two-spotted spider mite is an important pest of cotton and many other field crops. Early detection of plant damage caused by the mite is difficult because initial infestations tend to be scattered in small areas in the field. ARS researchers in College Station, Texas, adapted a hand-held light-reflecting (multispectral) instrument to detect the mite in growing cotton and beans. The instrument is capable of reliably distinguishing mite infested plants from non-infested plants, and is capable of differentiating between light, medium, and heavy spider mite infestations on cotton. This new technology will be very useful in detecting early spider mite infestations in cotton and other crops, and will guide rapid-response control procedures to assure effective protection of crops from mite damage; using the lowest amount of pesticide possible, with a minimum of adverse environmental impacts.

PERENNIAL CROPS:

Ring nematodes overcome resistance in grapevine rootstocks. The ring nematode is a common vineyard pest worldwide that reduces vine establishment and crop yields. The most cost-effective means to maintain vine productivity in nematode infested soils is to plant vines on nematode resistant rootstocks. ARS scientists in Corvallis, Oregon, showed that ring nematodes eventually reached high populations on two rootstocks (101-14 and 110R) that were previously found to be resistant in greenhouse trials, and that are in common use in the Pacific Northwest. Of six rootstocks tested, only 420A maintained no ring nematode population growth after 4 years in field microplots. These findings are particularly important for viticulturists in the Pacific Northwest, as 101-14 is a very common rootstock used in the region.

Continuous vine yield monitoring system. ARS scientists in Corvallis, Oregon, demonstrated that a continuous monitoring system in vineyard trellises, denoted the 'trellis tension monitor' (TTM) provided estimates of grapevine yields that were more accurate than those compiled by the juice processing industry using their static sample approaches. Scientists subjected the juice processors' data and the TTM data to three methods of calculating yield estimates. On average, across 10 commercial juice grape vineyards, TTM data produced more accurate estimates of actual yield than did the long-established, partially subjective protocols of the juice processors. Regardless of calculation method, there was high vineyard-to-vineyard variability in the accuracy of the yield estimate. TTM may be a useful adjunct to the traditional, labor-intensive practices that are required to estimate yield in vineyards; the TTM also provides nearly continuous information on the size of the crop, something that is not possible with the industry standard methods that rely on infrequent collections of fruit samples by hand.

Grapevines are far less vulnerable to water-stress induced formation of vapor bubbles than previously reported. Vulnerability to drought-induced formation of vapor bubbles was evaluated in grapevines using nuclear magnetic resonance in in-situ imaging of live grapevines to track movement of, and subsequent vessel blockage by, vapor bubbles. ARS scientists in Davis, California, revealed grapevines are not susceptible to significant drought induced vessel blockage within normal operating water potentials. The researchers also used High Resolution Computed Tomography, a type of CAT scan, to model the xylem network of grapevines. Using unprecedented resolution of this imaging technology, the researchers were able to describe the mechanism of air

bubble blockage repair in grapevines. Researchers worldwide had suspected this mechanism for decades, but had been unable to visualize this process in living plants.

Reducing Vineyard Weed Biomass. In the vineyard, pairing cultivation with glyphosate was as effective at reducing vineyard weed biomass as two glyphosate applications in 2 of 3 years, suggesting that substituting cultivation for one glyphosate application may be an effective method of reducing herbicide use in vineyards. ARS scientists in Davis, California, studied the effects on weed seedbank composition to determine potential long-term problems from using these practices. After 3 years, specific weed species – Carolina geranium, annual bluegrass, brome grasses, California burclover, and scarlet pimpernel – were associated with seedbanks of certain treatments. These do not pose problems with regard to the harvest of grapes, as such species are low-growing; suggesting that these practices will unlikely have strong interference with production.

Through implementation of no-till practices, vineyards may be a potential source of carbon sequestration. Due to the low level of soil disturbance in vineyards as compared to annual cropping systems like vegetables, vineyards represent a potential source for enhancing soil carbon pools in agriculture. ARS scientists in Davis, California, employed three management practices (no-till + cover crop, tillage + cover crop, and tillage + resident vegetation) and measured CO₂ emissions, soil water content, and temperature for 2 years. Effects of management practice interacted with climate and soil conditions before disturbance, i.e., management and rainfall, and influenced CO₂ emissions. Analyses of CO₂ emissions on gravimetric water content and soil temperature indicated that CO₂ emissions increased until water content reached 14 to 15 percent in tilled treatments and 20 percent in the mown treatment, subsequently declining; indicating thresholds of water content at which soil temperature more strongly influences CO₂ emissions. Findings suggest that no-till practices in vineyards may lead to greater soil carbon stabilization.

New technology to grow trailing blackberries without low-temperature injury. Trailing blackberries developed by ARS researchers in Corvallis, Oregon, are susceptible to low temperature injury and are not grown commercially in the central or eastern United States. A variety of techniques, such as rowcovers, cold frames, greenhouses, and high tunnels have been used to modify the growing environment for berry crops, but none has been shown to reduce winter injury in trailing blackberries. Researchers in Kearneysville, West Virginia, used a novel trellis system with a long rotating cross-arm (RCA) to position trailing blackberry canes close to the ground. Laying trailing blackberry canes close to the ground along with rowcover application is a cost-effective method for protecting trailing blackberries from cold temperatures and desiccating winds. This research enables trailing blackberries to be grown in the eastern United States. Since trailing blackberries ripen earlier than other blackberries they will be a useful variety for growers in the eastern United States interested in an early season blackberry. This research was awarded a technology transfer award from the Federal Laboratory Consortium, and the RCA trellis is currently being commercialized by Trellis Growing System, Inc., Fort Wayne, Indiana, which was awarded a Phase II USDA Small Business Innovation Research grant to expand blackberry production in the Midwest.

Blueberries form beneficial associations with ericoid mycorrhizal fungi. Blueberry plants form beneficial associations with ericoid mycorrhizal fungi (EMF), yet there is almost no information concerning how EMF influences the physiology of their host plants in horticultural production

systems. ARS researchers in Corvallis, Oregon, in collaboration with researchers at the University of Melbourne in Australia, compared the diversity and nutrient metabolism of EMF that exists in blueberry fields in Australia and the United States. Roots of blueberry plants from production fields in Australia contain fungi similar to EMF reported on northern hemisphere blueberry plants and on Australian native epacrids, which are related to the plant family for blueberry. These results indicate that information from research on EMF in Australian epacrids in natural environments can be useful for understanding the roles of EMF in blueberry production in the United States and Australia. Additionally, fungi in the same species varied their preferences for organic and inorganic forms of nitrogen indicating that different EMF may be more beneficial to blueberry plants grown in organic or conventional production systems.

Low nitrogen supply to Pinot noir increases berry flavor volatiles, but decreases yield and yeast available nitrogen. Nitrogen is likely the most important nutrient to manage in grapevine production, as it alters canopy and berry growth and influences the light environment around developing fruit clusters. Grape growers in western Oregon and Washington are interested in using low nitrogen supply to control canopy growth since vine access to soil water is so high early in the growing season. ARS researchers in Corvallis, Oregon, and Parma, Idaho, with collaborators at Oregon State University, discovered that reducing the nitrogen supply to grapevines grown in a sand-culture vineyard can increase the concentrations of certain flavor constituents and color compounds in berries, but that this effect of low nitrogen supply was not realized until crop yield, photosynthesis, and yeast available nitrogen in berries were below acceptable levels. These results suggest that reducing nitrogen supply to vines to improve berry quality parameters is not possible without sacrificing yield and berry yeast available nitrogen levels. Results from this research will help grape growers produce quality fruit without adversely affecting vine productivity and health.

Phytophthora root rot of blueberries. An ARS scientist in Poplarville, Mississippi, determined the relative susceptibility of 19 southern highbush blueberry cultivars to root rot and the effect of two cultural practices (raised beds and organic soil amendments) on the establishment of these cultivars in soils infested with the root rot pathogen. Most plants of each cultivar tested died within 4 years of transplanting, and the root rot pathogen was isolated from the roots of most of the plants that died. Neither cultural practice evaluated increased the survival percentage or vigor rating of any of the southern highbush blueberry cultivars. This information will be used by blueberry growers, extension specialists, and research scientists making planting recommendations for blueberries in areas where root rot is known to occur.

Brown marmorated stink bug can cause significant damage to apple and peach. The brown marmorated stink bug is a recently introduced invasive pest in the eastern United States that is a household nuisance and has begun to cause economic damage. ARS scientists in Kearneysville, West Virginia, showed that early season damage from the brown marmorated stink bug 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.

New and efficient mechanical thinning equipment for high density peach production systems. Reducing the number of fruit per tree (thinning) is necessary to attain marketable size fruit, but thinning is a costly and labor intensive operation that could benefit from mechanization. A new single spiked drum shaker was designed and developed by ARS scientists in Kearneysville, West Virginia, following previous tests with a double drum unit. The new shaker is smaller, lighter weight, and more readily adapted to forklift machines commonly found in commercial orchards. Field tests in 2010 demonstrated removal rates ranging from 10 percent to over 50 percent of the green fruit (about 35 days after full bloom) depending on shaker frequency. This thinning device has drawn interest from peach grower groups in California, Washington, and South Carolina, in addition to Pennsylvania where some tests were conducted.

Potassium deficiency can trigger excessive fruit-drop. The cause of unexplained severe fruit-drop in ‘Desirable’ pecan in commercial orchards was found by ARS scientists in Byron, Georgia, to be due to insufficient potassium in young developing fruit. Soil banding of potash rapidly increased the potassium concentration of foliage and fruit, and prevented fruit drop. Managing ‘Desirable’ foliage at a potassium concentration of 1.5 percent or more prevents potassium-associated fruit aborting in June. This research identified a deficiency in the potassium management recommendations by extension services, and sets new guidelines for orchard potassium management, as well as how to better improve potassium uptake from soils and how to more efficiently apply potassium to orchard soils.

Reducing alternate bearing in pecan trees by using gibberellic acid to increase tree resistance to black pecan aphids. Enhanced alternate bearing, and subsequent reduced profitability, is greatly accentuated by stress factors triggering premature senescence of canopy foliage or of premature defoliation. ARS Scientists in Byron, Georgia, found that treating canopies with gibberellic acid, a naturally occurring plant product, prevents black pecan aphids from triggering senescence of foliage or damage to tree canopies. However, the hormone can reduce return flowering if applied at the wrong time of the growing season or at an excessive concentration. Efforts are underway to optimize the approach for both aphid control and return flowering. The approach keeps populations of black pecan aphids from rising to damaging levels in orchards. This new tool has potential for use on all crops affected by senescence-inducing aphids.

Phosphite as a highly effective agent for controlling pecan scab in orchards. Premature fruit-drop or reduced kernel quality are devastating consequences of pecan scab disease on developing fruit. Additionally, there is emergence of scab resistance to certain classes of fungicides used in orchards. ARS scientists in Byron, Georgia, found that phosphite is highly efficacious for controlling scab in pecan orchards, although the mechanism of action is not yet fully understood. Certain phosphite products registered for use on other crops are now registered for use on pecan and are increasingly being used in commercial orchards to control pecan fruit scab.

Improving water use on pecan in the southwestern United States. Optimization of water use is a major goal in southwestern pecan orchards. A soil temperature model was developed by ARS scientists in Byron, Georgia, to predict the soil temperature where pecan roots grow; thus providing a means for estimating temperature impact on tree root health, and ultimately tree water use. The model potentially assists in efforts to further improve water use efficiency and minimize tree stress in pecan orchards.

Increasing pecan crop-load and yield with ReTain[®]. Excessive crop loss due to fruit-drop can greatly reduce profitability of certain orchard operations. A commercialized natural growth regulator product, AVG (ReTain[®]), was found by ARS scientists in Byron, Georgia, to possess potential for reducing nut-drop of certain pecan cultivars; thus increasing yield in “off” crop-load years. The research identifies ReTain[®] as a potentially useful horticultural tool for managing crop load on certain sensitive cultivars; thus, providing farmers with the means to improve nutmeat yield in “off” bearing years.

Leaf reflectance provides an estimate of sugarcane sucrose levels. Current methods used to estimate sugarcane stalk sucrose levels prior to harvest are labor and time intensive. ARS scientists in Houma, Louisiana, collected reflectance data from the leaves of predominant sugarcane varieties that were also sampled throughout two harvest seasons to determine sucrose accumulation (maturity). Leaf reflectance was effective at predicting sucrose in 36 to 79 percent of the cases, if varieties were combined; and in 65 to 100 percent of the cases if the varieties were considered separately. Several spectral regions were identified that appeared to be important in describing stalk sucrose levels, including: ultraviolet, blue, and green, and yellow, orange, and red, and the near-infrared wavelengths. These combined results indicate that it may be possible to utilize remote sensing techniques to estimate sugarcane maturity prior to harvest, which would allow growers and mills to more effectively manage field and varietal harvest schedules to insure maximum sucrose yields.

*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. The fungus causing this disease was described and illustrated by ARS researchers in Beltsville, Maryland, Mayaguez, Puerto Rico, and Hilo, Hawaii. The fungus’ relationship to other disease-causing fungi was also determined. This new knowledge will allow plant pathologists to accurately identify the cause of this disease, and is useful to plant regulatory officials working to control the spread of this disease.

*Identification and field assay of the pheromone of *Phyllophaga vandinei* and *Phyllophaga portoricensis*.* *Phyllophaga* species are important pests of fruit trees and turf. ARS researchers in Mayaguez, Puerto Rico, and collaborators, identified the pheromones of the two principal *Phyllophaga* pest species in Puerto Rico and reported these pheromones’ efficacy in attracting males for monitoring purposes. The pheromones may be used to monitor *Phyllophaga vandinei* and *Phyllophaga portoricensis* populations, eliminating the need for expensive and inefficient cone emergence traps. Studies are needed to determine the feasibility of mating disruption using these pheromones.

Rambutan resistance to acid soils. The most productive soils of the world are already under cultivation, and those available for agricultural expansion are often strongly acidic, possessing toxic levels of soil aluminum and/or manganese. These elements can drastically reduce crop yield when high concentrations are present in the soil. Incorporation of lime to the soil is a common practice to ameliorate acidity, but it is not very effective below the plough layer, and often lime is not available

to farmers with limited resources. ARS Researchers in Mayaguez, Puerto Rico, assessed the effect of soil acidity factors on dry matter production and leaf nutrient composition of four rambutan cultivars during a 2-year field study. High levels of soil acidity did not affect growth and dry matter production of rambutan seedlings, demonstrating that rambutan is highly tolerant to acidic soils, and that tolerance may involve a physiological process that keeps aluminum and manganese from entering the roots. Follow-up studies in nutrient culture confirmed tolerance to aluminum up to a concentration of 4.2 mM aluminum. The physiological mechanism of this tolerance, mainly exudation of organic acids, is being investigated. This study provides useful information to growers and extension personnel about the adaptability of rambutan to acidic soils.

GREENHOUSE, HIGH TUNNEL, AND NURSERY CROPS:

Critical boron and calcium tissue concentrations for pansy, petunia, and gerbera plugs. ARS scientists in Wooster, Ohio, investigated the cause of distorted plant growth in a series of experiments in cooperation with researchers at North Carolina State University. Boron deficiency was identified as the elemental problem which caused the stunted growth, thus eliminating calcium as an element of concern. Further investigation studied the causes of the deficiency in these small plants. This narrowed the primary cause of boron deficiency to be physiologically induced from over irrigation of plants with limited root substrate volume and a lack of environmental conditions which encouraged water uptake and transpiration. Through a series of presentations, scientific articles, and magazine articles, greenhouse operators are now aware of the steps required to better manage irrigation based on the transpirational needs of the plant; instead of reliance on timers which result in waterlogged conditions.

Visual symptoms of essential element deficiency for floriculture crops. Floriculture crops require a high degree of management to achieve market ready products. Fertilizer inputs are an important component of the production cycle, yet most floriculture crops lack critical tissue standards and adequate descriptions of the symptomology of nutritional disorders. In cooperation with scientists at North Carolina State University, ARS researchers in Wooster, Ohio, induced individual nutritional disorders in 14 floriculture crops. When incipient symptoms occurred they were recorded and photographed, and tissue samples were taken for analysis to determine nutritional levels. Through publication of results in both scientific journals and grower magazine articles and grower presentations, these nutritional evaluation criteria have been made available to the greenhouse industry to aid in the diagnosis of nutritional disorders.

Novel bi-layer hydrogel was developed for floriculture production. Floriculture production was in need of better means to mitigate the effect of biotic and abiotic stress induced by multi-day lack of soil hydration and nutrient availability. In cooperation with scientists at the University of Toledo, ARS researchers in Wooster, Ohio, developed a bi-layer hydrogel to separate the swelling requirement from that of low dehydration rates and mechanical strength. This would consist of a soft inner layer surrounded by a hard outer layer. The polyurethane coated gels produced the best results maximizing the water absorption of the inner layer while retaining sufficient compressive strength and reduced dehydration time of over a week. The development of these hydrogels could assist in not only more water efficient production strategies, but also in improved nutrient delivery in containerized plant production.

Water use of poinsettia cuttings in the propagation environment. In cooperation with Clemson University, ARS scientists in Wooster, Ohio, recorded gas exchange measurements, including photosynthesis, transpiration, and stomatal conductance, for poinsettia stock plants and on cuttings throughout the propagation environment. The data show that stomatal functioning is disturbed as a result of the removal of the shoot from the stock plant and function gradually returns to allow for more efficient water use as the cuttings are on the propagation bench. The differences are only observed during the night or when the cuttings experience water stress. As a result, the water use requirements are much higher on the first night in propagation as compared to the seventh night (before root initiation), even though the ability of the cutting to uptake water through the severed stem has not changed. Once root initiation takes place (between days 7 and 10), water use efficiency improved dramatically; however, photosynthesis did not return to the stock plant levels until the cutting was fully rooted in the fourth week of propagation. This information will help identify strategies to minimize water stress of newly propagated cuttings to improve rooting success.

Treatment with household bleach reduces Botrytis disease in cut rose flowers. Infection with the fungal pathogen *Botrytis cinerea* reduces the market value of many cut flower species. ARS scientists in Davis, California, showed that household bleach could reduce *Botrytis* infection on rose flowers when applied as a postharvest dip treatment. These results highlight the potential for household bleach use as a simple alternative postharvest treatment for the control of *Botrytis* on cut rose flowers. These findings also point to an exciting opportunity to test the effectiveness of this benign compound on additional rose varieties and flower species.

Thidiazuron (TDZ) extends the life of bulb flower plants. Early wilting and leaf yellowing are significant quality problems in a wide range of ornamental potted and bedding plants. Application of low concentrations of thidiazuron (TDZ) has been shown to be a very effective means of delaying leaf yellowing and flower senescence in alstroemeria, stock lilies, and cyclamen. ARS researchers in Davis, California, found that spray treatments with TDZ at the end of the production cycle, dramatically delayed leaf yellowing and extended flower longevity in a range of bulb flower plants, including tulips. These results indicate significant potential for TDZ as a tool to improve the postharvest life of bulb flower plants.

Effective methods for controlling pest insects in containerized production systems for ornamentals. Early detection of insect outbreaks is needed for effective chemical control of multigenerational pests, such as the strawberry rootworm beetle, a major root and leaf pest of potted azaleas in large production nurseries. Sticky traps are commercially available for monitoring strawberry rootworm beetles, but these cardboard traps are not durable enough to withstand daily application of irrigation water in a nursery. ARS researchers in Poplarville, Mississippi, have developed and tested a lightweight water-resistant trap that collects the strawberry rootworm beetle as well as other pest species. Incorporation of a light emitting diode increased trap catches by three- to seven-fold for the nocturnal strawberry rootworm beetle. Nursery managers now have a routine monitoring device for detecting early outbreaks of strawberry rootworm beetle and other leaf beetle pests. This station reduces the cost of labor and insecticides, as well as eliminates the need for the destructive sampling of plants.

Photosynthetic responses of important ornamental crops. Growers need information on what environmental factors affect photosynthesis of important floriculture crops to make environmental and lighting management decisions. This lack of information likely results in increased energy costs and/or reduced quality of yield. In a cooperative project with ARS scientists in Wooster, Ohio, and the University of Minnesota, the photosynthetic response of more than 20 important ornamental crops to light, temperature, and carbon dioxide was determined to develop environmental strategies to maximize photosynthesis that will lead to increased crop quality and yield. These data are being integrated into an ARS model (Virtual Grower) to allow application throughout the United States.

Environment influences efficacy of plant growth retardants. Plant growth retardant efficacy varies in different parts of the United States, between different growers, and between different application days. The basis for this variation is not understood and results in over and under application of these chemicals. ARS scientists in Wooster, Ohio, determined the impact of temperature and humidity on the efficacy of five plant growth retardants and identified that slow drying environments, at the time of spraying, results in the greatest efficacy. This information has been presented at multiple grower meetings and is being used to increase plant growth retardant efficacy to reduce chemical use.

Species suitable for northern U.S. green roofs identified. Installation of green roofs is a new strategy to reduce the environmental impacts of buildings, while reducing building energy costs. In cooperation with the University of Minnesota, ARS scientists in Wooster, Ohio, identified 42 plant species can survive on rooftops in the Twin cities area (a cold northern climate). A variety of desirable environmental impacts on each of those species was tested on a rooftop in Minneapolis and, could be used to reduce building operating costs. This information is being integrated into the Minneapolis planning departments building code requirement related to green roofs.

New heat- and/or drought-tolerant ornamental crops. Producers and consumers desire crops that can withstand heat and do not require irrigation. In collaboration with the University of Minnesota, ARS scientists in Wooster, Ohio, screened heat- and drought-tolerant species (over one hundred species) which have potential as new floriculture and/or landscape crops. The scientists selected 15 cacti species and five succulent species with significant potential as new crops. They also identified the flowering requirements of those crops to allow forcing as floriculture crops.

Lettuce seed quality enhanced by altering light and temperature during seed production. ARS scientists in Wooster, Ohio, in collaboration with Ohio State University scientists and the Ornamental Plant Germplasm Center in Columbus, Ohio, examined the influence of light quality and temperature in the maternal environment on the quality of lettuce seed. Uniform stand establishment is important for high yields. This study demonstrated that altering the red:far-red ratio of light during plant development and growing plants at higher temperatures resulted in seeds with greater germinability and longevity, providing a potential procedure for improved yield and quality of selected horticultural crops.

*X-ray technology used to assess quality of *Rudbeckia hirta* seeds.* Rapid assessment of seed quality is important in germplasm conservation. Traditional techniques such as chemical tests or germination can be time consuming. ARS scientists in Wooster, Ohio, in collaboration with

Brazilian scientists and the Ornamental Plant Germplasm Center in Columbus, Ohio, demonstrated that X-ray analysis could be applied to seeds of the popular ornamental plant *Rudbeckia hirta*. The results validated the use of this method to quickly assess the physical and physiological quality of *Rudbeckia* seeds.

Impact of composted dairy manure on pH management of soilless substrate. Substrate pH decline during cropping time is one of the greatest production challenges in containerized production. The use of dairy compost manure was evaluated for pH stabilization in containers by an ARS scientist in Wooster, Ohio, in collaboration with researchers at North Carolina State University. The scientists found that the pH buffering capacity of compost was similar to limestone. The potential exists of using a small amount of limestone in combination with compost to stabilize pH during crop production.

Nutritional contribution of mature dairy manure compost. In ongoing work with North Carolina State University and ARS scientists in Wooster, Ohio, mature dairy manure compost was used as a partial substitute for peat moss in a 3 peat moss:1 perlite mix used to grow an 11-week crop of pot mums. The crop tissue analysis at the end of the 11-week period indicated that compost resulted in higher leaf concentrations of potassium, sulfur, copper, iron, and manganese; lower, but adequate concentrations of calcium and magnesium; and similar concentrations of nitrogen, phosphorus, boron, and zinc. Results indicate that downward adjustments in fertilizer application are needed when using compost and that the adjustment is more important in the pre-plant fertilizer formulated into the substrate than in post-plant fertilization.

Impact of 13 floricultural crops on soilless substrate pH determined. Prevention of substrate pH shifts during crop production is a major challenge. A small body of evidence suggests that plant species differ in their impact on pH shift during production. ARS scientists in Wooster, Ohio, in collaboration with North Carolina State University, evaluated 13 floral crops for their effect on pH. The crops, when fertilized with neutral reaction fertilizer, lowered pH between 4 and 78 days after transplant by the following amounts: petunia (0.14), begonia (0.19), osteospermum (0.43), pansy (0.51), impatiens (0.79), New Guinea impatiens (0.89), geranium (0.97), vinca (1.00), tomato (1.17), Reiger begonia (1.46), pot mum (1.56), sunflower (2.44), and kalanchoe (2.45). The research showed that species clearly play an important role in pH shifts, and that crop species should be grouped according to their level of impact on substrate pH and the groups treated differently. Different fertilizers could be selected for each group according to the potential acidity/basicity rating of the fertilizers, thereby holding all groups within the same pH range during production.

Lime requirements of sphagnum peat moss determined from pH titration curves. A series of time consuming incubations of peat:lime mixtures are typically used to determine the liming rate required to achieve a desired pH. An ARS project by scientists in Wooster, Ohio, with North Carolina State University, developed a rapid, acid-based titration method for predicting lime requirement of sphagnum peat moss. This method was compared to the conventional method where peat was incubated with lime for 13 days in plastic bags until a steady-state was established. The two methods agreed with one another up to a pH of 6.2. Nevertheless, the acid-base titration is a quick method for accurately predicting lime requirements of acidic peat moss for target pH levels up to 6.2.

Energy-efficient management of greenhouse temperature for floriculture crops. Energy for heating is a significant expense in the production of greenhouse bedding plants. ARS scientists in Toledo, Ohio, in collaboration with scientists at Michigan State University, characterized how a variety of annual bedding plant species grow and develop in response to a range of greenhouse temperatures. Species-specific models were then generated that predict the effect of temperature on time-to-flower. Using that information, greenhouse growers can predict crop timing at different temperatures to meet market dates. Growers can use that information with the Virtual Grower computer program to identify production temperatures that consume the least amount of energy using their greenhouse characteristics and location. As a result, growers can potentially lower their energy bill by 10 to 20 percent or more and improve their scheduling accuracy. This information has been disseminated to greenhouse growers at regional and national grower meetings and through a 12-part “Energy-Efficient Annuals” series that appeared in Greenhouse Grower magazine in both print and on their Web site.

Virtual Grower En Español and Virtual Grower 2.6 were released. Growers of greenhouse crops face many complex, interrelated systems on a daily basis, wherein one decision influences others, sometimes in counterintuitive ways. Virtual Grower software developed by ARS scientists in Toledo, Ohio, allows users to build greenhouses with many different types of materials, select from different fuel types, simulate heating needs and costs, and see the impact of those decisions on the growth and development of some commonly grown greenhouse crops. The 2.6 version of the Virtual Grower program has added additional crops to existing capabilities to simulate supplemental lighting; the ability to add more fuels and materials for testing; improved calculations for energy curtains; and, real-time weather “calls” in which the next two days of weather at a grower’s U.S. site can be simulated. The software was translated into Spanish and is freely available for download at the ARS Web site. This software has assisted both large and small growers in identifying energy (and money) saving strategies in their operations, improved efficient scheduling of crops, and helped growers obtain energy efficiency improvement grants through various funding sources.

Substrate alternatives for high pH loving plants. Decreasing availability of traditional potted plant substrate materials such as pine bark makes it necessary to identify alternative substrate materials. Several different plants were grown in various blends of cedar with pine bark and sand. Plant response varied depending on the plant and the rate of cedar mixed into the substrate. The physical properties data showed that there was significantly less water-holding capacity and increased air space in the 80 percent cedar treatment. ARS scientists in Wooster, Ohio, attributed the differences in growth primarily to physical properties of the substrate rather than an allopathic response to chemicals in the cedar. The pH of cedar is 6.57 compared to that of pine bark at 4.17, which could make this a significant issue for plants requiring a low pH.

Transition of alternative switchgrass substrate from replicated research to adoption by nursery producers. Pine bark is currently used as the primary potting substrate for the nursery industry, but its cost is increasing, its availability decreasing, and it must be transported from the southern United States over long distances. ARS scientists in Wooster, Ohio, have developed a new potting substrate comprised primarily of switchgrass, a biofuel crop that can be grown and harvested locally. The goal was to apply the research findings to a commercial scale operation. The researchers used a cooperating commercial nursery’s equipment and traditional production

procedures to pot plants with the new substrate, and found that the substrate performed as well as the traditional pine bark-based nursery substrate. Successful adoption of switchgrass substrate could decrease the industry's reliance on pine bark that must be transported from far distances, and subsequently lower substrate cost.

Improved fertilizer practices for container-grown plants. Container-grown plants commonly exhibit a low recovery of nutrients from fertilizer. Fertilizer practices can be improved by understanding when and how plants most efficiently take up nutrients. Using container-grown woody and herbaceous perennial plants, including rhododendron, azalea, and hydrangea, ARS scientists in Corvallis, Oregon, in collaboration with researchers at Oregon State University and Mississippi State University, determined that the rate of nitrogen uptake is not proportionally related to uptake of other nutrients, and that nitrogen deficiency decreases a plant's ability to take up nutrients. Spraying foliage with urea in the autumn increases loss of certain nutrients from plants and increases demand for specific nutrients the following spring. These results can be used to improve efficiency of nutrient use and decrease fertilizer use in production of container-grown nursery crops by providing growers and fertilizer manufacturers with information on how to optimize fertilizer application based on rate, timing, method of nitrogen application, and cultivar.

Winter injury of bud and stem damage in nursery plants is linked to nitrogen management practices. Winter injury from cold, harsh weather decreases the quality of nursery trees and has a large impact on profitability of production systems. Observations linking winter injury of buds and stems to nitrogen management practices in nursery production are common. ARS scientists in Corvallis, Oregon, in collaboration with Oregon State University, Mississippi State University, and Washington State University, determined that both nitrogen rate and the form of nitrogen applied influenced autumn and winter cold tolerance of buds and stems on bareroot nursery trees. Also, researchers determined that trees with a similar nitrogen status withstood different levels of cold depending on the type or form of fertilizer being used. Consequently, fertilizer component selection may be as important for developing nutrient management strategies for nursery production of trees in climates prone to winter injury. These results can be used to modify nutrient management in nursery production systems to account for both productivity and end-product quality.

Autumn spraying alters tree metabolism. Spraying nursery trees in the autumn with defoliant and urea alter tree metabolism and could predispose trees to infection by pathogens, especially the pathogen *Pseudomonas syringae*, which causes large economic losses in nursery production in the Pacific Northwest. ARS researchers in Corvallis, Oregon, with collaborators at Oregon State University, determined the susceptibility of nursery trees to *Pseudomonas syringae*. Susceptibility is not the same in actively growing and dormant trees. Regardless of tree growth stage, environmental conditions after exposure to the pathogen, particularly temperature, are important for disease development. Additionally spraying trees in the autumn with urea, defoliant, or fungicides influenced tree nitrogen status and the effects on nitrogen status were unrelated to susceptibility to *Pseudomonas syringae*. These results can be used for developing practical recommendations for growers for integrating inspection and treatment protocols and optimizing autumn sprays to benefit both early harvest and prevention of the contamination and/or infection of *Pseudomonas syringae* in the field or storage.

SPRAY APPLICATION TECHNOLOGY:

Drift reduction technology evaluations. Drift associated with spray application of crop production and protection products is an ongoing concern for the spray application industry. An increasing number of drift reducing technologies are being developed and marketed for use with agricultural chemicals in aerial application treatments, but there have been no objective scientific assessments of the relative effectiveness of these various technologies in reducing spray drift. ARS researchers in College Station, Texas, developed and evaluated protocols for testing various drift reduction technologies in both high and low speed wind tunnels. This accomplishment will benefit both the agricultural application and regulatory communities because it provides objective, science-based information on the strengths and weaknesses of various drift reduction technologies. With increased demands placed on American farmers for higher crop production and from a changing climate, aerial applicators must ensure that each and every spray application provides the maximum benefit to the crop being sprayed. Appropriate utilization of these testing protocols will greatly support the commitment by all players in the aerial application industry to make applications in full compliance with all regulatory requirements and in an effective and environmentally sensitive manner.

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 prone to drift 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.

Automatically Triggered Multispectral Field Imaging System. An aerial multispectral imaging system was successfully employed on agricultural aircraft by ARS researchers in Stoneville, Mississippi, to determine areas of herbicide-induced crop injury after application of glyphosate. Field locations to image can be pre-programmed resulting in a hands-free, pilot-friendly imaging system. This multispectral platform shows promise as a new tool for operators of agricultural aircraft because imaging runs can be scheduled more frequently than conventional aerial remote sensing services. Frequent scheduling is particularly important when crop stress is being monitored. Convenience of this system could greatly increase the acceptance of remote sensing as a management tool by the agricultural community where agricultural aircraft are prevalent.

New application procedures increase herbicide efficacy. The wide use of the herbicide glyphosate (Round-Up[®]) on crops can create a situation where target weeds begin to develop resistance, thus making the herbicide less effective. When this happens, it can require applicators to use more and more glyphosate in their treatments, which increases costs to both applicators and farmers and

further exacerbates the weed resistance phenomenon. To address this growing problem, ARS researchers in College Station, Texas, studied how spray droplet size affects glyphosate effectiveness. The work showed that a higher spray droplet density (number of droplets per given area) results in better weed control. This knowledge allows applicators to adjust their application equipment in a manner to achieve good weed kill while using significantly less glyphosate and resulting in significant cost savings. An added benefit is that using less glyphosate to achieve the desired result also serves to slow down the process of weeds developing glyphosate resistance. Increasing the useful lifetime of an important and environmentally friendly herbicide like glyphosate also pushes into the future the need to shift to more expensive, environmentally harsh chemicals for weed control.

Drift reduction opportunities identified for tree crops. Orchard spraying can result in significant off-target spray movement if the spray delivery is not matched to the canopy development and the size of the canopy. ARS researchers in Wooster, Ohio, evaluated the differences in off-target spray movement using tower sprayers and conventional, axial-fan, air blast sprayers during dormant season and fully mature stages of canopy development. Significantly more material was detected above the canopy when the conventional orchard sprayer was used as compared to the tower sprayer. The dormant canopy permitted more material to flow through the target tree and deposit downwind, but a mature canopy deflected spray above the canopy where it could be carried away more easily by the wind. These results will help identify the off-target spray risks when fruit growers treat trees at different stages of development and help identify methods to keep more spray material in the target area.

Adjuvant effects on behavior of pesticide droplets on waxy leaves. Pesticide applications have ensured a bountiful supply of high quality crops, but the public has concern about increased use and effects on worker safety, environmental contamination, and adverse impacts on vulnerable ecosystems. ARS scientists in Wooster, Ohio determined residue patterns of single droplets and their evaporation time in four classes of adjuvants, on five different waxy plants, under controlled conditions. Results of this study demonstrated that the use of an appropriate class of adjuvants significantly improved droplet deposition on waxy leaves and the effectiveness of pesticides, and is critical to the process to reduce off-target losses.

Spray delivery parameters for effective wheat disease management. It is a challenge to protect cereal crops such as wheat from infection because different pathogens will cause infection in different parts of the plant canopy. The vertical (head and stem) and horizontal (leaf) structures may require different forms of treatment to achieve the necessary spray coverage for effective disease management. ARS researchers in Wooster, Ohio, evaluated the differences in spray deposits found on wheat heads, stems, and leaves following treatment by different droplet size nozzles and air-assisted delivery. Air-assisted delivery was found to be less effective at treating the upper canopy of the wheat. Twin-fan nozzles produced the highest deposits on the vertical plant surfaces (heads and stems). Growers and pest management specialists can use this information to optimize set-up of their wheat plant protection equipment to protect different parts of the canopy from infection, resulting in higher crop yields.

A real-time variable-rate sprayer for nursery liner applications. Pesticide applications on ornamental nursery tree liners with conventional sprayers for control of pests and diseases are

excessive. ARS scientists in Wooster, Ohio, developed a real-time, variable rate experimental sprayer – where spray outputs coincided with canopy sizes – to reduce pesticide usage. In laboratory tests, the experimental sprayer reduced the average application rates by 59 to 75 percent and demonstrated that pesticide usage could be greatly reduced when variable rate spray applications are determined by canopy sizes. The potential use of this variable rate sprayer could be extended to other fruit crops, including vineyards.

Pesticide spray delivery to mature poinsettias. Few recommendations are available on how to best match application systems to greenhouse ornamental production systems. ARS researchers in Wooster, Ohio, evaluated foliar pesticide deposit characteristics in a mature poinsettia canopy when applied by handgun and boom delivery systems. All systems tested had difficulty treating the underside of leaves in the poinsettia canopy. Air-assisted, boom delivery produced the most uniform deposits, which could result in more predictable insect control. Droplet size decreased with increasing nozzle pressure in the handgun systems, but air entrainment aided delivery in the handgun applications as well as the air-assist boom systems. Producers can use these findings to better match the pest management need with the pesticide application options they have available, as well as reduce overall pesticide use and improve pest control.

Improved weed management for potted nursery plants. Herbicide delivery through a potted nursery plant to the surface of the pot is difficult because the canopy can intercept a significant portion of the spray material. ARS researchers in Wooster, Ohio, using potted hydrangea plants and various application tools which have been shown to provide improved canopy penetration in field crop applications, including large droplet nozzles, air-assisted delivery, and increased spray volume applications, showed that nearly 50 percent of the spray material was captured by the plant canopy and unavailable to provide weed control on the surface of the pot. The multi-port air-assist sprayer produced the highest canopy deposits but the lowest spray deposits on the pot surface. There were no significant differences in the deposits on the pot surface for coarse spray as compared to medium spray nozzles. Spray volume had a slight effect but depended on the overall droplet size. There were no significant differences in droplet size. Commercial potted plant producers can use this information to better target the pot surface with weed control materials and more accurately estimate the amount of herbicide which will be available, so that they can provide the desired weed control.

Component 2: Bees and Pollination

HONEY BEES [*APIS*]:

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 and is in commercial production under the name HopGuard®.

Comparative genomics reveals vital information about insect immunity. Honey bees combat American foulbrood, viruses, and *Nosema* disease with immunity proteins found across insect species. An ARS bee scientist in Beltsville, Maryland, helped define those immune proteins in wasp and aphid genomes, allowing a broad comparative look at bee immunity and possible targets for breeding programs. These efforts, along with experimental studies of honey bees in the field, indicate that bee immunity is also aided by colony level defenses, including the use of protective plant resins and hygienic behaviors that remove infected brood.

Commercialization of varroa-resistant honey bees selected for pollination performance. Varroa mites are an external parasite of honey bees and the major cause of colony losses throughout the United States. Bees with varroa sensitive hygiene (VSH), which have good resistance to varroa mites, were tested by ARS researchers in Baton Rouge, Louisiana, for two seasons in a commercial migratory beekeeping operation focused on crop pollination. Bee colonies were created from VSH queens which were outcrossed, i.e., matings were not controlled, a method used by most large-scale beekeepers. Bee colonies were shipped nationwide and used for spring pollination of almonds in California, apples in New York, low-bush blueberries in Maine, and cranberries in Massachusetts, as well as late summer honey production in New York. VSH bee colonies performed well in terms of survival, populations, and resistance to varroa mites. The best surviving VSH bee colonies from each year were propagated to form a breeding population which had enhanced genetics for both mite resistance and behavior related to crop pollination. These bees are now being marketed by a Cooperative Research and Development Agreement partner (Glenn Apiaries), and the use of their germplasm should improve adoption of mite-resistant bees by commercial beekeepers that pollinate crops.

Hybrid Russian bees show promise for control of tracheal mites. Because bee stocks in the United States have only been exposed to tracheal mites since their introduction here 25 years ago, they have not had time to develop resistance to these mites. However, Russian honey bees evolved with the tracheal mite and have excellent resistance, although the genetic control of the resistance was not understood. From analysis of recently imported tracheal mite resistant Russian queens and susceptible queens, ARS researchers in Baton Rouge, Louisiana, determined that tracheal mite resistance is dependent on a number of genes with major dominance interacting with minor genes. Beekeepers can benefit from this resistance to tracheal mites by either using pure Russian queens or queens resulting from outcrosses of Russian queens, the latter requiring less work for stock maintenance.

NON-APIS BEES:

Possible link between exposure to fungicides and reduced honey bee health revealed. Honey bee colonies store pollen as bee bread (microbe processed pollen), which is their source of protein. ARS scientists in Tucson, Arizona, found that bees fed pollen contaminated with fungicides had a lower diversity of microbes in their stored pollen than those fed pollen without fungicides. Worker bees from colonies with fungicide contaminated pollen had a lower protein content compared with those fed uncontaminated pollen. Reduced protein content is a symptom of poor digestion and

causes colonies to reduce the size of their brood. Adult workers with low protein titers have reduced immune response and longevity. Research findings therefore suggest that fungicides might compromise the ability of workers to process pollen which could negatively affect colony nutrition and therefore imperil growth and survival.

Publication of the symptoms of colony collapse disorder. Satisfying an objective of the colony collapse disorder (CCD) action plan, ARS researchers in Beltsville, Maryland, concluded a descriptive epidemiological study describing the level of pests and other pathogens present in CCD versus non-CCD colonies. This information was used to define the symptoms of CCD. The scientists' research documented high levels of pathogens in colonies suffering from CCD and suggested that CCD is an interaction between pathogens and other stress factors. While not demonstrating causation, this prioritizes the areas of research that should be conducted to address uncertainties in bee health related to CCD.

Plants broadly attractive to bees needed in restoration of rangelands damaged by fires. Wild bee communities provide pollination services critical to large post-fire restoration projects in rangelands, but their fates following fires were largely unknown. ARS scientists in Logan, Utah, found that fires in rangeland plant communities that were in good shape before burning did not destroy native bees because the bees nest below ground; allowing them to rebound the next year. However, wildflowers which the bees depend on may be gone after a fire, and without them the bees cannot survive. This work demonstrates the importance of initial seeding in restoration projects. Initial seedings should include plants that are broadly attractive to local native bees in order to sustain these surviving bees until the native plant communities can re-establish, which can take several years. This work will be used by land managers in restoring habitat.

Nesting methods successfully established for four bumble bee species. Bumble bees are important pollinators of commercial greenhouse crops, but are difficult to rear in captivity. Because it is particularly difficult to get queens to establish new nests in captivity, ARS scientists in Logan, Utah, tested three nest establishment methods on four species of bumble bees. Results show that some techniques greatly increased the success rate (four-fold) in some species, in comparison to other techniques. By targeting proper rearing techniques to a given species, producers and researchers will be able to save time and resources when producing bumble bees in captivity.